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Research and Development

EPA

Zinc

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ZINC

by

Subcommittee on Zinc
Committee on the Medical and Biologic Effects of
Environmental Pollutants
National Research Council
National Academy of Sciences
Washington, D.C.

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The project that is the subject of this report was approved by the Governing Board of the National Research Council, whose members are drawn from the Councils of the National Academy of Sciences, the National Academy of Engineering, and the Institute of Medicine. The members of the Committee responsible for the report were chosen for their special competences and with regard for appropriate balance.

This report has been reviewed by a group other than the authors according to procedures approved by a Report Review Committee consisting of members of the National Academy of Sciences, the National Academy of Engineering, and the Institute of Medicine.

FOREWORD

The many benefits of our modern, developing, industrial society are accompanied by certain hazards. Careful assessment of the relative risk of existing and new man-made environmental hazards is necessary for the establishment of sound regulatory policy. These regulations serve to enhance the quality of our environment in order to promote the public health and welfare and the productive capacity of our Nation's population.

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To aid the Health Effects Research Laboratory to fulfill the functions listed above, the National Academy of Sciences (NAS) under EPA Contract No. 68-02-1226 prepares evaluative reports of current knowledge of selected atmospheric pollutants. These documents serve as background material for the preparation or revision of criteria documents, scientific and technical assessment reports, partial bases for EPA decisions and recommendations for research needs. "Zinc" is one of these reports.

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The Introduction was written by Dr. Robert I. Henkin, Chairman of the Subcommittee. Chapter 2, on properties and uses of zinc, was written by Mr. Carl H. Cotterill. Dr. Michael Fleischer contributed Chapter 3, on natural sources and distribution of zinc, except for the section on zinc reactions with organic matter in soils, which was prepared by Dr. Bernard D. Knezek.

Except for the passage on sewage in the section on waste disposal, which was written by Dr. Knezek, Chapter 4 was the responsibility of Dr. Jerome F. Cole. Chapter 5, on zinc in plants, was the work of Dr. Knezek, who was solely responsible for the section on aquatic plants, and jointly responsible with Dr. John F. Davis (since deceased) for the section on terrestrial plants.

Drs. Douglas A. Wolfe and Bruce R. Stillings prepared the account on zinc in aquatic animals, set forth in Chapter 6. Chapter 7, on zinc in humans, was written by Dr. Henkin, and Chapter 8, on zinc in the diet and the effects of zinc deficiency in animals, was written by Dr. Jean Apgar. Dr. Joseph E. Coleman prepared Chapter 9, on zinc in metalloproteins. Chapter 10, on clinical aspects of zinc metabolism, was the work of Dr. Henkin. Three authors were involved in the preparation of

Chapter 11, on toxicity of zinc; Dr. Magnus Piscator wrote two sections, those on zinc and cadmium and interactions between zinc and cadmium, and Dr. Apgar wrote the section on animals. Dr. Robert A. Goyer had overall responsibility for the chapter, and contributed the other sections.

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Ms. Avis Berman edited the manuscript and worked tirelessly with the authors in resolving the many difficulties that arose during the preparation of the voluminous typescript. Ms. Joan Stokes checked for accuracy all references cited, and was responsible for preparation of the extensive bibliography.

Free use was made of the resources available at the National Library of Medicine, the National Agricultural Library, the Library of Congress, and the Air Pollution Technical Information Center of the Environmental Protection Agency. Also acknowledged is the assistance given to the Subcommittee by the National Research Council's Advisory Center on Toxicology, the National Academy of Sciences Library, the Environmental Studies Board, and various units of the National Research Council.

CONTENTS

CHAPTER 1

INTRODUCTION

Zinc has been used by man for industrial, ornamental, or utilitarian purposes for nearly 2,000 years, but its history is clouded until well into the middle ages. Thompson[1608] was able to ascribe the capability of making a compound of copper, tin and zinc by reduction with charcoal to the Babylonians of 3000 B.C.

Semitic bronzes found at Gezer in Palestine and dated 1400 and 1000 B.C. contained up to 23% zinc as well as 10% tin.[1211] Although brass was known early in Palestine and was probably used for cymbals and bells, zinc was probably quite unknown. The brass was likely to have been made by smelting a zinc ore with copper and charcoal.

The oldest known piece of zinc is the form of an idol found in the prehistoric Dacian settlement at Dordosch, Transylvania. Analysis of the idol showed that it was composed of 88% zinc, 11% lead, and 1% iron.[1025] In the ruins of Camiros, destroyed in 500 B.C., two bracelets filled with zinc were found;[1025] in the ruins of Pompeii, destroyed in 79 A.D., the upper part of a fountain front was discovered, and the finders claimed that it had been covered with zinc.

The Greeks may also have known about and used zinc. Aristotle was said to have spoken about a brilliant white copper produced by adding "some kind of earth" rather than melting tin with copper.[1042] Theophrastus, a contemporary of Aristotle, also mentioned an ore which became superior in beauty and color when mixed with copper.[1042] Strabo (60 B.C.-20 A.D.) described an ore "which when burned becomes iron and when heated in furnaces with a certain earth distils 'mock silver' (pseudargyros), and this with the addition of copper makes the 'mixture'.....named oreichalkos."[1042] Mitchell concluded that

pseudargyros was zinc and the method of smelting through distillation was one which would produce zinc. Other historians are not convinced that pseudargyros was metallic zinc and suggest that it might have been metallic arsenic or arsenical copper.[1025]

It is not clear when zinc was introduced as a useful agent in the Far East. However, useful brass products were made by the calamine* process certainly since the beginning of the Christian era,[53] and perhaps even before it.[12] Li suggested that zinc was used by the early Chinese by about the beginning of the Christian era.[264] Forbes speculated that zinc metal was prepared by Indian alchemists in the laboratory in the twelfth century, but the process was not applied industrially.[489] The Chinese also were said to know how to make a metal from tutty scraped from the sides of smelting furnaces making lead. The substance was thought to be cadmium, but it was mostly zinc oxide.[370]

The Romans were probably the first Europeans to make brass when they began to use it for coins (about 20 B.C.). They discovered that when copper was smelted with calamine ore, a yellow alloy more gold than bronze could be obtained.[718]

In its isolated form, zinc was not recognized until the fifteenth century when smelting probably occurred accidentally.[12] The word "zinc" may be derived from the German noun Zinker, meaning "jagged part or tooth." This word was applicable to the metal because in early smelting endeavors,

*A zinc ore.

zinc was deposited in the furnace in the form of pointed parts. Upon slow cooling, zinc vapor will condense and form clusters of hexagonal crystals with pointed ends. "Spelter" was also a name given to zinc and it is related to the word "pewter." The earliest occurrence of the word "spelter" was in 1661.

Commercial smelting began in the eighteenth century when it was clearly realized that zinc could be obtained from the calamine used to make brass and that it was the same metal imported from India and China.

No reports of zinc toxicosis in any form were forthcoming from these early accounts. However, it has been alleged that zinc was applied as an ointment for skin lesions by several cultures of the ancient world, including the Egyptians and other Mediterranean peoples. The first documented usage of zinc administered orally occurred in 1826 when zinc sulfate was used to treat gleet and leucorrhea.[554]

The purpose of this document is to attempt to place into perspective the role of zinc in the environment -- its importance as an essential nutrient for all forms of life and its effects as a toxic agent to some species. The effects of zinc are many sided. Because zinc is abundantly distributed throughout the earth's crust and found in many manufactured products, humans come into direct daily contact with several forms of zinc. Zinc is used to manufacture motor oils, lubricants, and rubber tires, and it is found in the fuel oil and coal used for heating and manufacturing purposes; thus, particulate zinc is present in the atmosphere in rural as well as urban areas. This particulate zinc has not been specifically identified as a public health problem. Zinc is essential for the normal activity of DNA polymerase; and hence, for protein synthesis. It plays an important role in the growth, development, and metabolism of living species. In plants and animals the balance between excess and insufficient zinc is important. Plants do not grow well in zinc-depleted or zinc-absent soils, and insufficient dietary zinc in animals and humans leads to poor

development and other pathophysiologic changes. Dietary zinc replacement usually will reverse the pathologic events of zinc depletion in man and animals, but important exceptions do occur. In rat offspring with congenital malfunctions or behavioral abnormalities associated with zinc depletion, repletion with zinc seems to be of little value. In humans, a major problem is the diagnosis and evaluation of zinc deficiency, particularly if the case is marginal.

Excessive zinc in the aquatic environment is of particular importance, because the respiratory systems of fish are very sensitive to the toxic effects of zinc. Water represents a convenient vehicle for disposal of human and animal waste products, which can be quite high in zinc. The toxic effects of zinc in humans are not common medical problems, although they may appear in some metal workers and in some children under special conditions. There is increasing evidence that zinc may be helpful in the treatment of several disorders of man and animals, including skin lesions and the rare disease acrodermatitis enteropathica. However, knowledge of zinc metabolism in humans is still quite limited.

CHAPTER 2

PROPERTIES AND USES OF ZINC*

Among the major common metals (iron, aluminum, copper, lead and zinc), all but zinc are easily recognized as such by the consumer. This distinction is a function of its loss of identity to an end product. Zinc has been found to be beneficial and necessary for proper nutrition of humans, animals, and plants, and a deficient intake has been proved deleterious.

Zinc is a bluish-white, relatively soft metal with a density slightly less than iron (7.133 and 7.86 g/cc, respectively). Its atomic number is 30, atomic weight, 65.37, and it is placed in Group II-B of the periodic table. The atomic radius of zinc is 1.31 Å and its electron configuration is 2-8-18-2. Zinc is divalent in all its compounds. It is a composite of five stable isotopes: zinc-64, -66, -67, -68, and -70; measured by relative abundance, these constitute 48.86%, 27.62%, 4.12%, 18.71%, and 0.69%, respectively, of the whole. Six radioactive isotopes have been identified: zinc-62, -63, -65, -69, -72, and -73.

The most commonly used artificial isotopes are zinc-65 and zinc-69, which have half-lives of about 244 days for the zinc-65, 14 h for the zinc-69 isomer, and 58 min for zinc-69 itself. Decay products of zinc-65 and -69 are stable copper-65, manufactured by positron emission and electron capture from zinc-65, and stable gallium-69 isotopes, produced by negative

*Much of this chapter is derived from the U.S. Department of Interior, Bureau of Mines publication, The U.S. Zinc Industry: A Historical Perspective (1974).[1016] No other references will be provided in the body of the chapter unless the information comes directly from another source.

β-emission from the zinc-69 isotopes. Other characteristics of zinc isotopes may be found in Weast *et al.*[1740]

The structure of the zinc molecule exhibits a hexagonal close-packed lattice: in Å, when a = 2.664 and c = 4.9469, a/c = 1.856.

PHYSICAL PROPERTIES OF ZINC

Cast zinc, which crystallizes in a hexagonal system, is brittle; but when heated to about 120 C, it becomes ductile and is easily rolled or drawn. After mechanical shaping at about this temperature, the metal does not become brittle again upon cooling. Thus, wrought zinc is used in construction to form roofing and as drawn battery cans for dry power cells. The high electrochemical activity of zinc is surpassed among the common metals only by magnesium and aluminum in the electromotive series. This activity accounts for one of its major uses: the galvanizing of iron and steel. In such applications, zinc undergoes sacrificial corrosion from the surface of the steel, and protects the substrate from structural degradation. Zinc readily combines with other metals, imparting characteristics of workability at low temperature, corrosion resistance, and pleasing finishes for use in die-casting alloys, brass, and other common alloys. Table 2-1 sets forth some physical properties of zinc.

CHEMICAL PROPERTIES OF ZINC

Pure zinc at ambient temperatures is highly resistant to attack by dry air, but in temperatures above 225 C the rate of corrosion increases rapidly. In moist air, attack proceeds at room temperature, and in the presence of carbon dioxide it accelerates to form the hydrated basic carbonate, $2ZnCO_3 \cdot 3Zn(OH)_2$. This carbonate

TABLE 2-1

Some Physical Properties of Zinc[a]

Density:

solid at 25 C, 7.133 g/cm^3
solid at 419.5 C, 6.83 g/cm^3
liquid at 419.5 C, 6.62 g/cm^3
liquid at 800 C, 6.25 g/cm^3

Melting point: 419.5 C (692.7 K)

Boiling point (1 atm): 907 C (1,180 K)

Heat capacity:

solid - Cp = 5.35 + 2.40 x 10^{-3} T (298 - 692.7 K) cal/mol
liquid - Cp = 7.50 cal/mol
gas (monatomic) - Cp = 4.969 cal/mol

Heat of fusion: 1,765 cal/mol at 419.5 C

Heat of vaporization: 27,430 cal/mol at 907 C

Linear coefficients of thermal expansion:

polycrystalline (20 - 250 C), 39.7 x 10^{-6} per C

Volume coefficient of thermal expansion (20 - 400 C): 8.9 x 10^{-5} per C

Thermal conductivity:

solid (18 C) 0.27 cal/sec cm C
solid (419.5 C) 0.23 cal/sec cm C
liquid (419.5 C) 0.145 cal/sec cm C
liquid (750 C) 0.135 cal/sec cm C

Modulus of elasticity:

10 to 20 x 10^6 psi

Surface tension (liquid): γ = 758 - 0.09(t - 419.5 C) dynes/cm

Electrical resistivity:

polycrystalline (t = 0-100 C) R = 5.46(1 + 0.0042t) microhms/cm^3
liquid (423 C) 36.955 microhms/cm^3

Magnetic susceptibility (diamagnetic):

polycrystalline (20 C) - 0.139 x 10^{-6} cgs electromagnetic units

[a] Modified from Schuhmann and Schadler,[1451b]

forms a tightly adhering light gray film which tends to protect the zinc from further corrosion. Halogens react with zinc in the presence of moisture, but not under dry conditions. Reaction with steam at 350 C or higher occurs rapidly. Mineral acids also easily attack zinc: the strongest reaction occurs with sulfuric acid, followed by hydrochloric, and nitric acids. Zinc displays a vigorous reducing power, liberating hydrogen from sulfuric and hydrochloric acids. This property is the basis for the use of zinc dust or mossy zinc in many commercial organic chemical processes. Zinc liberates nitrogen oxides instead of hydrogen from nitric acid. The metal is amphoteric, reacting with hot caustic to liberate hydrogen and form zincates. All zinc is inert to petroleum derivatives and anhydrous alcohol but is oxidized by mixtures of alcohol and water. The chemical properties of zinc compounds make them useful as oxides, carbonates, sulfates, sulfides, chlorides, phosphates, and organic complexes. Commercial grades of zinc contain enough trace impurities to make it more reactive than pure zinc.

ZINC METAL AND ALLOYS

Commercial grades of zinc metal have been established by the American Society of Testing Materials (ASTM), as listed in Table 2-2.

TABLE 2-2

Grades of Slab Zinc and Chemical Requirements[a, b]

Grade	Composition, %			
	Lead, maximum	Iron, maximum	Cadmium, maximum	Zinc, minimum by difference
Special High-Grade[c]	0.003	0.003	0.003	99.990
High-Grade	0.07	0.02	0.03	99.90
Intermediate	0.20	0.03	0.40	99.5
Brass Special	0.60	0.03	0.50	99.0
Prime Western	1.60	0.05	0.50	98.0

[a]Data from ASTM standards, specification B6-70.[30a]
[b]When specified for use in the manufacture of rolled zinc or brass, aluminum shall not exceed 0.005%.
[c]Tin in Special High-Grade shall not exceed 0.001%.

Prime Western was the specification earliest established for use in hot-dip galvanizing. Brass Special and Intermediate were largely used in alloying with copper to form brass. High-Grade, and eventually Special-High-Grade specifications were established for zinc to be used in alloys containing small amounts of aluminum. These alloys are used in the die-casting method of producing intricate functional parts cast to very close dimensional tolerances.

Basic alloying characteristics are governed by the maximum solid solubilities of other metals in zinc; for example, gold is soluble to the extent of 10-15%, silver, 8%, cadmium, palladium, and copper, 2-3%, manganese, 0.5-1%, and aluminum, 3-5%. In forming intermetallic compounds, higher percentages prevail. The effects on the physical and chemical properties of zinc by lead, cadmium, iron, tin, copper, aluminum, magnesium, and titanium in either the solid solubilities or intermetallic compound forms are described below.

Lead. Small quantities of lead in rolled zinc used for manufacture of dry cells promotes a desirable rate of chemical reaction for the proper release of electrochemical energy.

Cadmium. In rolled zinc, cadmium tends to increase strength, hardness, creep resistance, and recrystallization temperature.

Iron. Alloys of iron and zinc in the substrate of galvanized steel are of paramount importance to a properly galvanized article. A cross section

of the coating shows first a layer of pure iron overlaid by a very thin layer of $FeZn_3$, over which is a thicker layer of $FeZn_7$, above which is a layer of essentially pure zinc with a few solid crystals of the $FeZn_7$. These various alloys are quite resistant to corrosion and form an adherent bond between the steel base and the zinc. If the galvanized sheet is to be subsequently bent and formed, it is necessary to minimize the alloy formation to avoid cracking and peeling.

Tin. Tin is deleterious in very small quantities in rolled zinc, because it causes ruptures during hot rolling. In castings it also promotes subsurface corrosion. However, small amounts of tin will cause an esthetically desirable "spangle" on galvanized sheets.

Copper. Copper increases strength, hardness, creep resistance, and recrystallization temperature. It may bring on corrosion of zinc in dry cells.

Aluminum. At levels of 3.5-4.5%, aluminum reduces grain size, and improves impact strength and castability of zinc. In galvanizing, die casting, and protective galvanic anodes, aluminum beneficially inhibits formation of the zinc-iron alloy.

Magnesium. Magnesium counteracts subsurface corrosion effects of tin and lead in zinc alloys.

Titanium. Titanium forms a compound rich in zinc. At elevated temperatures, it decreases the grain size of cast zinc and restrains grain growth in rolled zinc. Used in newer rolled zinc applications, it greatly increases creep resistance.

One of the chief uses for high-purity zinc is in zinc-base alloys for die-casting. The die-casting process enables a dimensionally accurate equipment part to be produced in a die-casting machine in fractions of a minute. Therefore, few finishing operations are required before use, and the economic advantages are obvious. Table 2-3 reports the chemical and physical properties of die-cast alloys.

TABLE 2-3

Chemical and Physical Properties for Zinc Die-Casting Alloys[a]

Chemical Requirements as Ingot[c]	Alloy #3 (ASTM AG40A)	Alloy #5 (ASTM AC41A)	Alloy #7[b]
Copper, %	0.10 (max)	0.75-1.25	0.10 (max)
Aluminum, %	3.9-4.3	3.9-4.3	3.9-4.3
Magnesium, %	0.03-0.06	0.03-0.06	0.005-0.020
Iron, % (max)	0.075	0.075	0.050
Lead, % (max)	0.005	0.005	0.0020
Cadmium, % (max)	0.004	0.004	0.0020
Tin, % (max)	0.002	0.002	0.0010
Nickel	---	---	0.005-0.020
Zinc	remainder	remainder	remainder

Chemical Requirements as Alloy Die-castings[d]	Alloy #3 (ASTM AG40A)	Alloy #5 (ASTM AC41A)	Alloy #7
Copper, %	0.25 (max)	0.75-1.25	0.25 (max)
Aluminum, %	3.5-4.3	3.5-4.3	3.5-4.3
Magnesium, %	0.03-0.08	0.03-0.08	0.005-0.020
Iron, % (max)	0.100	0.100	0.75
Lead, % (max)	0.007	0.007	0.0030
Cadmium, % (max)	0.005	0.005	0.0020
Tin, % (max)	0.005	0.005	0.0010
Nickel	---	---	0.005-0.020
Zinc	remainder	remainder	remainder

[a] Alloys are numbered 3, 5, and 7 in ordinary trade practice.
[b] Alloy 7 has not been assigned an ASTM number.
[c] ASTM standards, specification B240-64.
[d] ASTM standards, specification B86-71.

TABLE 2-3 continued

Typical Mechanical Properties for Die-Casting Alloys	Alloy #3 (ASTM AG40A)	Alloy #5 (ASTM AC41A)	Alloy #7
Charpy test impact strength, ft-lb, 1/4 x 1/4-in. (.625 x .625 cm) bar, as cast	43	48	40
Charpy test impact strength, ft-lb, 1/4 x 1/4-in. bar, after 10 yrs indoor aging	41	40	---
Tensile strength, psi, as cast	41,000	47,600	41,000
Tensile strength, psi, after 10 yrs aging	35,000	39,300	---
Elongation, % 2 in. (5 cm), as cast	10	7	14
Elongation, % 2 in. after 10 yrs aging	16	13	---
Expansion (growth) in./in. (2.5 cm/2.5 cm) after 10 yrs aging at room temperature	0.00008	0.00007	---
Brinell hardness	82	91	76

Die-cast alloys are used widely in producing automobile parts such as carburetors, grills, door handles, and ornaments; appliance control panels, home washer parts,television bezels, transistor radio and camera frames; and small control gears and many other devices. A zinc-containing alloy in growing use is an aluminum-based die-casting alloy incorporating 2.7-8% zinc. Production with this alloy requires higher die temperatures and slower operating speeds than necessary for the zinc-base alloy, but parts made of it exhibit tensile strengths up to 80,000 psi. Many solders, especially those used on aluminum, contain zinc as well as other alloying metals. Silver and gold solders also are alloyed with zinc.

Zinc is a minor component of the copper-zinc alloys known as brass (although brass is the oldest known use of zinc). Commercial brasses contain between 5-40% zinc. Brass is superior to copper alone in its greater strength and ductility, and its resistance to corrosion. It is widely used in hardware, plumbing accessories, instruments, communication equipment, as well as for aesthetic purposes because of its pleasing yellowish-gold color. Table 2-4 lists composition and properties of some of the more popular commercial brasses.

Wrought zinc is composed of commercial grades of zinc to which very small amounts of other metals have been added to alter the properties of the zinc. Important properties of these wrought zinc alloys designed for commercial use include: resistance to corrosion; white and nonstaining corrosion products; chemical characteristics desirable for dry cells and photoengraving plates; mechanical properties for easy forming, machining, and spinning; and good solderability. Table 2-5 lists several of these zinc alloys, and their composition, characteristics and uses.

TABLE 2-4

Properties of Common Brasses[a]

Material	Nominal Composition, % copper	zinc	other	Condition	Yield Strength, 10^3 psi	Tensile Strength, 10^3 psi	Brinell Hardness	Density, g/cc	Modulus of Elasticity, 10^6 psi
Gilding metal	95	5	-	Cold-rolled	50	56	114	8.90	17
Commercial bronze	90	10	-	Cold-rolled	54	61	125	8.80	17
Red brass	85	15	-	Cold-drawn	55	70	120	8.75	17
Aluminum brass	76	22	2[b]	Annealed	27	60	82	8.33	16
Admiralty brass	71	28	1[c]	Annealed	20	53	60	8.53	16
Cartridge brass	70	30	-	Cold-rolled	63	76	155	8.47	16
Yellow brass	65	35	-	Cold-drawn	55	70	115	8.47	15
Nickel silver	65	17	18[d]	Cold-rolled, hard temper	70	85	170	8.75	18
Muntz metal	60	40	-	Annealed	20	54	80	8.39	15
Manganese bronze	58.5	39.2	1,1,0.3[e]	Cold-rolled	50	80	180	8.36	15
Architectural bronze	57	40	3[f]	Annealed	20	60	95	8.47	14

[a] From the Chemical Engineers' Handbook.[1234]

[b] Aluminum

[c] Tin

[d] Nickel

[e] Iron, tin, and manganese, respectively.

[f] Lead

TABLE 2-5

Classification of Wrought Zinc Alloys[a]

Composition %						
Lead	Iron	Cadmium	Copper	Other	Characteristics	Typical use
0.05–0.10	0.012 max	0.005 max	0.001 max	-	High ductility with low hardness and stiffness. Very little work hardening possible.	Drawn battery cans, eyelets, fuse links, and many other industrial articles drawn, formed, and spun.
0.05–0.10	0.012 max	0.06	0.005 max	-	High ductility with low hardness. Can be work hardened slightly.	Drawn battery cans, eyelets and grommets. Extruded battery cans. Address plates, laundry tags.
0.15–0.35	0.017 max	0.15–0.30	0.005 max	-	High hardness and stiffness. Uniform etching quality. Can be work hardened.	Photoengraver's plates, lithographer sheets, boiler and ship plates, weatherstrips.
0.05–0.10	0.012 max	0.005 max	0.85–1.25	-	High hardness and stiffness. Good ductility. Good creep resistance. Work hardens easily.	Weatherstrips and drawn and formed industrial articles requiring stiffness.
0.05–0.10	0.015 max	0.005 max	0.85–1.25	0.007–0.02[b]	High stiffness and creep resistance. Can be severely work hardened.	Flat or formed commercial articles requiring high stiffness and strength.
0.005–0.10	0.012 max	0.05 max	0.50–1.50	0.12–1.50[c]	Outstanding creep resistance. Can be severely work hardened. Lowest thermal expansivity with the grain. Very high resistance to grain growth during annealing.	Corrugated roofing, leaders and gutters, and other uses requiring maximum creep resistance.
0.15–0.35	0.014–0.025	0.15–0.30	0.005 max	0.005–0.025[b]	High hardness. Can be baked without serious softening. Good etching characteristics.	Photoengraver's sheets.
0.007 max	0.10 max	0.007 max	0 – 3.5	0.02–0.10[b]; 3.5–4.5[d]	High strength and hardness.	Shearing and forming dies. Extruded rods, tubings and moldings.

[a] From the Metals Handbook.[959]

[b] Manganese.

[c] Titanium.

[d] Aluminum.

Although not a strict alloying function, the enlistment of zinc as galvanizing to coat iron and steel objects is a major use of the metal. As mentioned, dipping steel in a bath of molten zinc results in a coating of 1-2 oz zinc/ft^2 (311.1-622.2 g/m^2) of steel surface. This coating galvanically protects the underlying steel from corrosion because of the higher electromotive potential of zinc over iron. Thus the zinc coating must be entirely corroded away before the iron substrate begins to rust, and the structural strength of the article is preserved by the sacrificial corrosion of zinc. Steel dipped in zinc is the material of construction for galvanized garbage cans, barbed and woven wire fencing, steel woven cable, highway guard rail, radio and electricity transmission towers, and building structures and bridges. Recent advances in continuous-line galvanizing have made possible better control of the composition of minor alloying metals in the zinc bath possible. The newer processes are able to provide superior, more economical coatings on continuous steel sheet made for protective building siding, downspout and guttering, air ducts, and automobile and appliance bodies.

INORGANIC ZINC COMPOUNDS

Because of its high reactivity with other elements and amphoteric character, zinc forms a wide variety of compounds. Zinc sulfates and chlorides are water soluble, whereas the oxides, carbonates, phosphates, silicates, and organic complexes generally are insoluble. Properties of common zinc compounds are tabulated in Table 2-6. In the following discussions of zinc compounds, substances will be listed in generally descending order of commercial volume.

TABLE 2-6

Properties of Common Inorganic Zinc Compounds[a]

Zinc Compound	Formula	Molecular Weight	Crystal Form	Refractive Indexes	Specific Gravity	Melting Point, C	Boiling Point, C	Remarks
Oxide	ZnO	81.37	Hexagonal	2.008, 2.029	5.606	1,975	-	
Sulfide (sphalerite)	ZnS	97.43	Cubic	2.368	4.102	-	-	Transition point @ 1,020 C
Sulfate	$ZnSO_4$	161.43	Orthorhombic	1.658, 1.669	3.54	600	-	
Chloride	$ZnCl_2$	136.28	Trigonal	1.681, 1.713	2.91	283	732	Forms hydrates
Ammonium chloride	$ZnCl_2.2NH_4Cl$	243.26	-	-	1.8	-	-	
Fluoride	ZnF_2	103.37	Tetragonal	-	4.95	872	1,500	
Bromide	$ZnBr_2$	225.19	Trigonal	-	4.01	394	650	
Iodide	ZnI_2	319.18	Trigonal	-	4.7364	446	624	
Acetate	$Zn(C_2H_3O_2)_2$	183.46	Monoclinic	-	1.84	200	-	
Borate	$3ZnO.2B_2O_3$	383.35	Triclinic	-	4.22	980	-	
Carbonate	$ZnCO_3$	125.39	Trigonal	1.818, 1.618	4.398	-	-	Evolves carbon dioxide @ 300-500 C
Chromate	$ZnCrO_4$	181.36	Orthorhombic	-	3.40	-	-	
Dichromate	$ZnCr_2O_7.3H_2O$	335.40	-	-	-	-	-	Hygroscopic
Cyanide	$Zn(CN)_2$	117.41	Cubic	1.470	1.852	800	-	
Nitrate	$Zn(NO_3)_2$	189.40	-	-	-	-	-	Unstable; forms hydrates
Orthophosphate	$Zn_3(PO_4)_2$	386.05	Monoclinic	-	3.998	900	-	Forms hydrate and complex
Orthosilicate	Zn_2SiO_4	222.82	Trigonal	1.694, 1.723	4.103	1,509	-	
Fluosilicate	$ZnSiF_6.6H_2O$	315.54	Trigonal	1.382, 1.396	2.104	-	-	Decomposes in water

[a]Data from the *Handbook of Chemistry and Physics*.[1740]

Zinc Oxide

Zinc oxide, one of the most valued zinc compounds, crystallizes in a white hexagonal form. Combustion control during formation can modify the particle size and shape to achieve desired properties. A variety of particle shapes and wide size ranges combine with the large interstitial spaces of the compound to provide a kaleidoscope of properties to be manipulated. Zinc oxide is used most in the compounding and vulcanizing of rubber, wherein the properties of high heat capacity and conductivity serve to cool flexing rubber in belts and tires. This compound scavenges any free sulfur remaining in the article after the rubber is vulcanized.

The high index of refraction of zinc oxide accounts for its use as a pigment in white paints to bestow high hiding power. For exterior paints, the ability of a thin film of zinc oxide to completely absorb ultraviolet rays from the sun is useful; it also acts as an effective mildewcide and prevents fungal staining.

Zinc oxide commonly is classified by method of production. American Process zinc oxide is made by carbon reduction of roasted zinc ore, and the resultant zinc vapor is burned to form zinc oxide; French Process zinc oxide is produced by burning zinc metal in air; some chemical process oxide is made by precipitating zinc hydroxide ($Zn[OH]_2$) from solution and calcining it to form oxide.[1607c] ASTM specifications and chemical and physical test values for typical grades of zinc oxide are set forth in Table 2-7, although it should be noted that many producers manufacture many more grades than are listed to accommodate certain specific needs and uses.

TABLE 2-7

ASTM Specifications and Physical and Chemical Test Values for Zinc Oxide

A. ASTM Specification for Zinc Oxide[a]

	Zinc Oxide	
	American Process	French Process
Zinc oxide, range or minimum %	98	99
Total sulfur, maximum %	0.2	0.1
Moisture and other volatile matter, maximum %	0.5	0.5
Total impurities, including moisture and other volatile matter, maximum %	2.0	1.0
Coarse particles (total residue retained on a No. 325 [44 μm] sieve , maximum %)	1.0	1.0

[a] ASTM standards, specification D79-44 (reapproved 1974).[31]

B. Chemical and Physical Property Test Values for Representative Grades of Zinc Oxide[b]

	Rubber Grade	Paint Grade
Chemical tests, in %		
Zinc Oxide	99.20	99.20
Lead	0.03	0.03
Cadmium	0.01	0.01
Manganese	0.003	-
Copper	0.002	-
Chlorine	-	0.02
Acidity as sulfur trioxide	0.03	0.15
Total sulfur as sulfur trioxide	0.05	0.20
Water-soluble salts	0.15	0.20
Insoluble in hydrochloric acid	0.15	0.15
Loss at 110 C	0.30	0.30
Physical tests		
Average numerical diameter, 1 μm	0.28	0.26
Average surface diameter, 3 μm	0.65	0.60
Specific surface, m^2/g	1.69	1.83
% fines under .50 μm	16.0	18.0
% through 325-mesh/in^2 (6.5 cm^2) screen	99.97	99.92
Specific gravity	5.65	5.65
Apparent density, lb/ft^3 (kg/m^3)	30.0 (220)	30.0 (220)
Oil absorption of zinc oxides, g oil/100 g zinc oxide	-	14.5

[b] A "typical" chemical and physical analysis of one company's particular grades of rubber and paint zinc oxides.

Zinc oxide is insoluble in water, organic solvents, and neutral oils. With organic and inorganic acids, it forms simple and complex salts and soaps. Reaction with alkalies form zincates, because zinc is amphoteric. The oxide may be used to catalyze some chemical reactions. One of the oldest uses is pharmaceutical -- in treatment of burns, infections, and skin diseases. A newer use for zinc oxide, which relies on physicochemical properties, was taken up by the photoconductivity field: carefully produced zinc oxides are coated on paper and used in office photocopying applications. The ceramic industry produces frits and glazes in which zinc oxide provides color for pottery or improves the brilliance of glass.

Other Inorganic Zinc Compounds

Zinc sulfate. Zinc sulfate, $ZnSO_4$, is water-soluble and useful as a hardener in viscose rayon spinning baths, and as a flotation reagent in mineral concentrations. It has become increasingly popular as a trace element applied to overcome zinc deficiencies in plants grown in certain areas.

Zinc chloride. Zinc chloride, $ZnCl_2$, has a low melting point and is used in galvanizing fluxes, and in preserving and controlling inflammability in wood. It is also an essential ingredient in dry cells, a disinfectant, a printing mordant, and an aid for mercerizing cotton.

Zinc sulfide. Zinc sulfide, ZnS, is usually a component of barium lithopone, a white paint pigment. Very pure zinc sulfide enjoys widespread use as a phosphor in cathode-ray television tubes and fluorescent lamps.

Zinc chromate. Zinc chromate, $ZnCrO_4$, is a wood preservative, an algicide, and a primer on metal surfaces for protection against corrosion.

Zinc carbonate. Zinc carbonate, $ZnCO_3$, like zinc oxide, is now being used more and more as a nutritive supplement for swine, sheep and poultry.

Zinc borates. Zinc borates, $3ZnO.2B_2O_3$, are used as fire retardants and as fluoborates for insecticides.

Zinc acetate. Zinc acetate, $Zn(C_2H_3O_2)_2$, chiefly provides mordant for dyeing and glazes for porcelain, but it is also an astringent and antiseptic.

Zinc silicate. Anhydrous zinc silicate, Zn_2SiO_4, is used as a phosphor in television screens.

Zinc fluosilicate. Zinc fluosilicate, $ZnSiF_6.6H_2O$, has continued to be an effective laundry sour, concrete hardener, and wood preservative.

Zinc cyanide. Zinc cyanide, $Zn(CN)_2$, serves as zinc carrier in electroplating, and it is used for medicinal purposes to treat epilepsy, neuralgia, etc. It is poisonous, and will evolve hydrogen cyanide, HCN, gas if contacted by mineral acids.

Zinc nitrate. Zinc nitrate, $Zn(NO_3)_2$, finds use as a mordant in dyeing.

Zinc phosphate. Zinc phosphate, $Zn_3(PO_4)_2.4H_2O$, mostly is used in dental cements.

Zinc phosphide. Zinc phosphide, Zn_3P_2, is a common ingredient in rat and mouse poisons. It is dangerous to humans if highly toxic phosphine gas liberated.

Zinc permanganate. Zinc permanganate, $Zn(MnO_4)_2.6H_2O$, finds some application as an antiseptic and astringent.

Zinc peroxide. The powdered form of zinc peroxide, ZnO_2, functions as a deodorant, astringent, and antiseptic for wounds and skin diseases.

ORGANIC ZINC COMPOUNDS

Most organic derivatives of zinc are manufactured in very small commercial quantities, whereas many of the inorganic compounds are sold in lots of thousands of tons. As in the previous section, the list of compounds is in approximate descending order of commercial volume.

Zinc soaps. Zinc soaps--stearates, $Zn(C_{18}H_{35}O_2)_2$, palmitates, $Zn(C_{16}H_{31}CO_2)_2$, and oleates, $Zn(C_{18}H_{33}O_2)_2$--are fatty acid salts. They are fine, bulky, soft, white powders that feel greasy, repel water, and generally are soluble in benzene and petroleum derivatives. They are used as lubricants and mold release compounds in rubber and plastic forming, metal die-casting, and medicine tablet making. Zinc soaps also waterproof concrete, paper, and textiles, and serve as flatting agents in lacquers.

Zineb and ziram. Zineb (ethylenebis; $C_4H_6N_2S_4Zn$) and ziram (dimethyldithiocarbamate; $C_6H_{12}N_2S_4Zn$) are popular agricultural fungicides, because they are more tolerable to humans than are the mercury, lead, and copper fungicides which the zinc salts replace. Some plants even reap nutritional value from the zinc.

Zinc bacitracin. Zinc bacitracin is an antibiotic in ointments and preparations for human use and a growth stimulator in swine and poultry nutrition.

Zinc propionate and caprylate. Zinc propionate, $Zn(C_3H_5O_2)_2$, and caprylate, $Zn(C_8H_{15}O_2)_2$, serve as fungicides on adhesive tape coatings, and preparations which combat athlete's foot and other molds, fungi, and bacteria.

Zinc phenolsulfonate. Zinc phenolsulfonate, $Zn(C_{12}H_{10}O_8S_2 \cdot 8H_2O)$, is both an insecticide and an antiseptic for internal treatment of ulcers and wounds.

Zinc salicylate. Zinc salicylate, $(C_7H_5O_3)_2Zn.3H_2O$, is an effective astringent and antiseptic.

Many other organic salts are used in medicinal applications because small quantities of zinc are toxic to many microorganisms harmful to human, animal, and plant life.

Zinc undecylenate. Zinc undecylenate, $C_{22}H_{38}O_4Zn$, is used as a cutaneous fungicide to control dermatophytoses.

PATTERNS OF USE

In the United States, the use of zinc in all forms has increased at a rate approximating that of the real growth of the gross national product. The pattern of use has shifted from one in which brass and galvanizing were predominant to one in which a much larger share is constituted by zinc-base alloys for die-casting applications. Consequently, the proportion of zinc used for brass, wrought zinc, and other purposes has decreased. Table 2-8 and Figure 2-1 chart some of these trends from 1935-1976. During these years, the total zinc usage in the United States has more than tripled from 566,000 to 1,753,000 metric tons in 1973. This total for 1973 includes the most frequently used statistic of "slab zinc" consumption (1,364,000 metric tons), plus the zinc content of ores used directly without going through the cast metal or slab stage (118,000 metric tons), as well as the recoverable zinc content of alloys and chemical compounds produced from old and new scrap, residues, etc. (271,000 metric tons).

During the period charted, the four major uses of zinc appear to have been at or near their peak in 1973 (except for brass, used heavily during World War II for cartridge cases). These large applications of zinc are very sensitive to the economic cycle (note 1975 data), because zinc is important in automobile production (die-casting, galvanizing, and oxide for rubber tires) and industrial and residential construction activity (galvanizing, brass, die-casting for appliances, and oxide for paint pigments).

TABLE 2-8

Total Zinc Usage in the United States

Classified by Industry, at Intervals between 1935 and 1976[a]

	Metric Tons						
Year	Zinc Base Alloy	Galvanizing	Brass	Oxide	Rolled Zinc[b]	Other	TOTAL
1935[c]	58,468	176,901	139,706	90,598	51,256	49,318	566,247
1940	137,639	300,464	249,403	105,677	54,608	58,818	906,609
1945	125,847	305,886	441,013	125,768	88,531	77,087	1,164,132
1950	277,973	400,691	272,850	140,478	62,091	71,071	1,225,154
1955	413,367	409,268	270,790	126,633	46,801	65,868	1,332,727
1960	322,576	337,100	187,284	101,542	35,104	67,765	1,051,371
1965	602,386	437,645	258,467	145,388	41,623	94,868	1,580,377
1970	444,535	430,232	262,904	158,572	37,254	92,231	1,425,728
1973	576,493	511,504	343,210	189,950	36,980	94,476	1,752,613
1975	312,516	341,906	236,172	120,048	24,773	82,069	1,117,484
1976[d]	460,900	390,600	256,900	123,000	27,200	35,400	1,294,000

[a] Data from U.S. Department of Interior, Bureau of Mines.

[b] Included in "Other" on Figure 2-1.

[c] Partially estimated.

[d] Estimated.

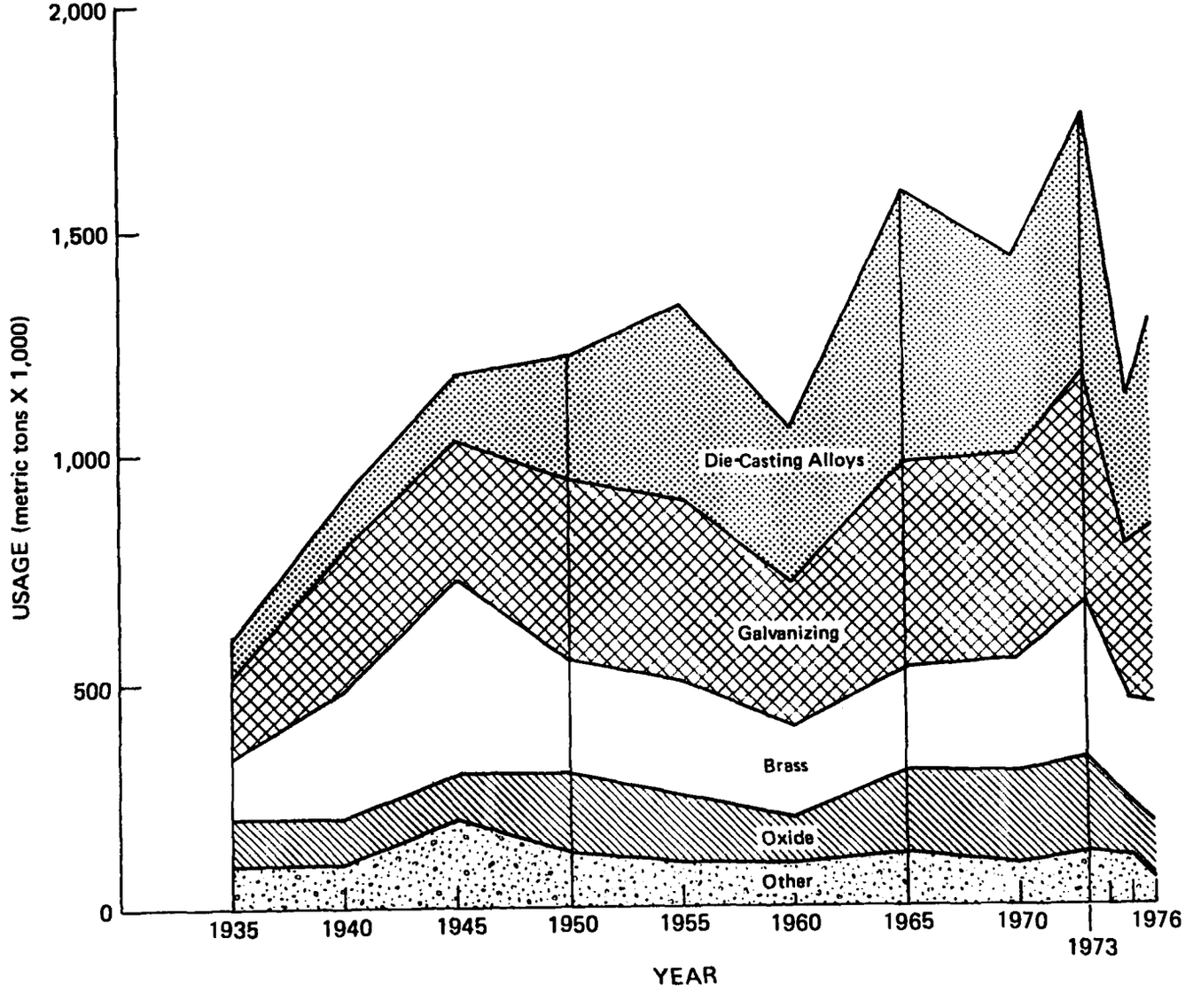


FIGURE 2-1 Total zinc usage in the United States by industry at selected intervals, 1935-1976. Compiled from data of U.S. Department of Interior, Bureau of Mines.

CHAPTER 3

NATURAL SOURCES AND DISTRIBUTION OF ZINC

Zinc is a moderately abundant element; its concentration in the continental crust of the earth is generally given as 70 ppm, which would place it as twenty-fourth in abundance of the chemical elements.

Zinc has ionic radii of 0.68Å in fourfold coordination and 0.83Å in sixfold coordination; they are close to those of magnesium, ferrous iron, and cupric copper, and these elements commonly form solid solutions with zinc in oxygen salts such as the sulfates and phosphates.

Although more than 80 zinc minerals are known, only a few serve as commercial ores of the metal. The principal ores are the sulfides sphalerite and wurtzite (cubic and hexagonal ZnS) and their weathering products, especially smithsonite, $ZnCO_3$, and hemimorphite, $Zn_4Si_2O_7(OH)_2 \cdot H_2O$. Other minor ores are zincite, ZnO, and willemite, Zn_2SiO_4.

Zinc is present in part in igneous and metamorphic rocks as the sulfide sphalerite, but most of it present is disseminated as a minor constituent of rock-forming minerals, especially those rich in iron, such as magnetite, Fe_3O_4, the pyroxenes $(Mg,Fe)_2Si_2O_6$ and $Ca(Mg,Fe)Si_2O_6$, the amphiboles, such as $Ca_2(Mg,Fe)_5Si_8O_{22}(OH)_2$, biotite, $K(Mg,Fe)_3AlSi_3O_{10}(OH,F)_2$, spinel, $(Mg,Fe)Al_2O_4$, garnet, $(Fe,Mg)_3Al_2(SiO_4)_3$, and staurolite

$(Fe,Mg)_2Al_9Si_4O_{23}(OH)$. When igneous and metamorphic rocks are weathered, most of the zinc is concentrated in the clay minerals of the sedimentary rocks and soils formed, especially in the minerals of the montmorillonite group.

ZINC IN IGNEOUS AND METAMORPHIC ROCKS

Ranges of zinc content and averages for various types of igneous rocks are assembled in Table 3-1. Zinc generally is concentrated in basaltic rocks and is somewhat depleted in granitic rocks. In basaltic rocks, the content of zinc increases directly with the total iron content.

Data on metamorphic rocks indicate that their contents of zinc are very similar to those of the unmetamorphosed rocks in which they were found, that is, no indication of appreciable mobilization of zinc during metamorphism has been found.

ZINC IN SEDIMENTARY ROCKS

Ranges of zinc content and averages for various types of sedimentary rocks are listed in Table 3-2. Zinc is concentrated notably in shales and clays; part of it is thought to be an isomorphous replacement for magnesium and iron and part may be adsorbed on the surface. The concentration is clearly higher in black shales rich in organic matter,[1627,1694] but the nature of the zinc-organic complex and the exact mechanism of its formation are not known. Zinc exhibits a two- to threefold concentration in iron-rich laterites and bauxites.[791] Jenne has reviewed evidence indicating that hydrous oxides of manganese

TABLE 3-1

Zinc in Igneous Rocks[a]

Type of Rock	No. of Analyses	Range, ppm	Average, ppm
Ultramafic	85	25-103	55
Basaltic	1,681	42-420	100
Intermediate	114	5-127	70
Granitic	1,087	5-235	50
Rhyolitic and dacitic	300	15-400	48
Alkalic	252	18-1,070	70

[a]Derived from Wedepohl, 1743a Gurney and Ahrens, 587a and 1366 Rosman.

TABLE 3-2

Zinc Contents of Sedimentary Rocks[a]

Type of Rock	No. of Analyses	Range, ppm	Average, ppm
Limestones and dolomites	490	< 0.1- 180	20
Sandstones	150	5 - 170	30
Shales low in bituminous matter	365	46 - 200	95[b]
Shales high in bituminous matter	980	15 -1,500	200
Phosphorites	240	20 - 750	100[c]
Coal	1,600	7 -1,000	40-80

1743a

[a] Derived from Wedepohl.

[b] But the average for deep sea clays was about 165 ppm.

[c] Appreciably higher (average, 300 ppm zinc) in phosphorites of the Phosphoria formation of Montana, Wyoming, Idaho, and Utah.[575]

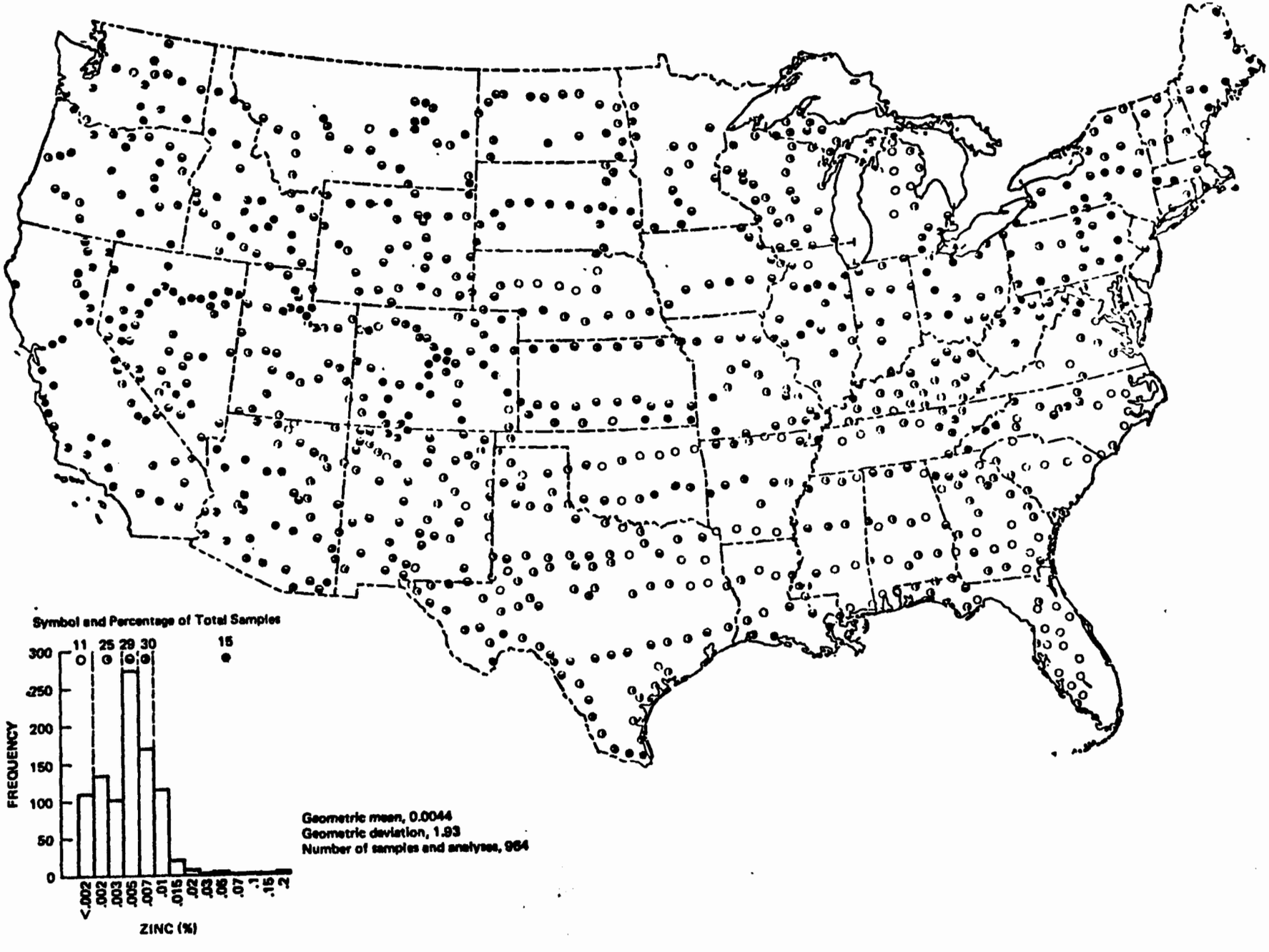
and iron are the principal controls of the fixation of zinc in soils and fresh water sediments.

Zinc is also quite concentrated in marine phosphorites, and especially high levels of zinc are reported in those from Idaho, Montana, Wyoming, and Utah.[575] These high levels of zinc also are found in phosphatic fertilizers manufactured from phosphorites of this area.

ZINC IN SOILS

The zinc content of soils has been the subject of some review.[153,938,1581,1696] Normal soils contain 10-300 ppm zinc, average about 50 ppm, and in uncontaminated areas, the contents generally are not very different from those of the parent rock. A recent study of 863 U.S. soils[1477a] noted an average of 54 ppm zinc (see bar graph for Figure 3-1). Zinc contents of soils near sulfide deposits are generally higher than background; this indicator is the basis of geochemical prospecting methods widely used in exploration for ore deposits.

Soils near highways may be contaminated appreciably; zinc contents of such soils[891] are set forth in Table 3-3. The source of the zinc may be from wearing of tires (containing zinc oxide) and emissions from motor oil to which zinc dithiophosphite has been added; these sources of zinc are in addition to industrial emanations. Klein[851] found that soil from industrial areas near Grand Rapids, Michigan, had a mean content of 56.6 ppm zinc, whereas nearby agricultural and residential areas had respective means of 22.1 and 21.1 ppm zinc. Davies found similar conditions in England.[359]



1477a

FIGURE 3-1 Zinc in soils of the continental United States. Reproduced from Shacklette et al.

TABLE 3-3

Zinc Content in Soils near Highways[a]

Location	Meters from Road	ppm Zinc Extractable by 1 N Hydrochloric Acid Depth of soils, cm 0-5	5-10	10-15
Near U.S. 1, Beltsville, MD	8	172	94	72
"	16	66	48	42
"	32	54	46	42
Baltimore-Washington Parkway, Bladensburg, MD	8	162	86	36
"	16	110	28	20
"	32	44	20	18
West of Interstate 29, Platte City, MO	8	54	24	16
"	16	60	21	16
"	32	15	11	14
North of Seymour Road, Cincinnati, OH	8	72	24	11
"	16	60	16	10
"	32	34	11	8.3

[a]Data from Lagerwerff and Specht.

ZINC REACTIONS WITH ORGANIC MATTER IN SOILS

Many organic substances, including humic and fulvic acids and a wide range of biochemical compounds, form stable combinations with zinc.[711,1435,1553] The occurrence of insoluble zinc-organic matter complexes has been established, and the formation of soluble organic complexes with zinc has a profound effect on the mobility and availability of the metal. The soluble zinc-organic complexes can leach through the soil to influence weathering and geochemical distribution in the soil profile. Organic substances are also important in the transport and ultimate concentration of zinc and other metals in such important deposits as peat and coal. Some soluble zinc-organic complexes are so stable that the zinc is essentially unavailable to living systems, but these extremely stable complexes are rare. Therefore, the chemistry of the zinc-organic complex in soil is closely related to the nature and magnitude of microbial and other biologic activities as well as to the level of zinc and other competing compounds present.

Natural Chelating Substances in Soil

Two main groups of organic compounds that form stable compounds with zinc in soil are biochemicals which exist in living organisms, and complex polymers formed by secondary synthesis reactions to form more complex products. The first group contains organic acids, peptides, proteins, and polysaccharides, and the second group contains humic and fulvic acids. The two groups cannot always be clearly separated because some biochemical compounds are bound tightly to humic materials. Zinc can function as a linkage in binding these organic compounds, and it can be chelated by them. Most of the insoluble zinc-organic complexes are associated with the humic group, especially humic acid.

The most soluble and least stable zinc complexes are with individual biochemical compounds; but the fulvic acid complexes also have high water solubilities, yet greater stability than the biochemical complexes.[1553]

Biochemical Compounds

The relative susceptibility of simple biochemical compounds to microbial decomposition has often been assumed to be of little or no importance in metal reactions in soils. But recent studies show that the combined total of potential chelating agents at any one time may tremendously influence the availability of zinc and other metals to plants.[526,713] For example, up to 75% of the zinc in displaced soil solutions was shown to be associated with low molecular weight, dialyzable organic constituents.[526,713] Natural complexing biochemicals have also been demonstrated to be of considerable importance in the transport of zinc to plant roots and its movement through the soil profile.[425]

Many organic acids (malic, citric, oxaloacetic, fumaric, α-ketoglutaric, pyruvic, etc.) and free amino acids (alanine, aspartic, glutamic, glycine, cystine, cysteine, etc.) are the biochemical compounds of primary importance in the formation of soluble zinc complexes in soils. Other compounds that chelate zinc in soils include organic phosphates, phytic acid (inositolhexaphosphoric acid), chlorophyll and chlorophyll-degradation products, simple sugars, porphyrins, phenolic compounds, and auxins. The contribution of all these other compounds to zinc-organic complexes from biochemical sources is minor compared to the prevalence of organic acids and free amino acids.[711,1081] Specific reactions have been reviewed in detail.

Humic and Fulvic Acids

The binding of zinc and other metals by humic and fulvic acids is influenced greatly by the nature and properties of the acids. The following differences should be noted between fulvic acid and humic acid:

- fulvic acid is light yellow to yellow-brown, whereas the color of humic acid ranges from dark brown to gray-black;
- the degree of polymerization is increased for humic acid;
- the molecular weight of fulvic acid is about 2,000, and the molecular weight of humic acid is about 300,000;
- carbon content increases from 45% in fulvic acid to 62% in humic acid;
- oxygen content decreases from 48% in fulvic acid to 30% in humic acid; and
- exchange acidity decreases from 1,400 in fulvic acid to 500 in humic acid.[1553]

Generally, humic acid is the material extracted from soil by alkaline solutions and precipitated upon acidification, whereas fulvic acid is an alkaline-soluble material that remains soluble after acidification.[1553]

Formation of stable humic and fulvic acid complexes with zinc and other metals is possible because of the high content of oxygen-containing functional groups such as carboxyl, phenol, alcohol, enol-hydroxyl, and carbon-oxygen structures. Amino and imino groups may also be important in zinc binding.[1553] Several functional groups have been identified[1314] as being involved in the binding of zinc by humic acids. Phenolic hydroxyl and carboxyl groups having pK$_a$ values between 2.8 and 4.4 accounted for the least stable but greatest adsorbed fraction of zinc, while more stable fractions were attributed to strongly acidic carboxyl groups with pK$_a$ values below 2.0. Zinc would be more likely to be adsorbed by the more stable forms.

Randhawa and Broadbent[1314], Schnitzer and Skinner[1436], and Hodgson[711] have suggested that humic and fulvic acid fractions of organic matter are very important in zinc adsorption. Randhawa and Broadbent[1314] found that the species of zinc complexed by the humic acid varied with pH. At pH 7.0, they concluded that 70% of the metal retained by humic acid was present as the divalent species, whereas at pH 3.6, 75% of the acid was monovalent. However, inherent errors in determining the ratio of the complexes by the ion exchange resin method may cast doubt on their conclusions. Stability constants for the zinc-humic acid complex were calculated to be 4.42 at pH 3.5, 6.18 at pH 5.6 and 6.80 at pH 7.0.[1315]

Schnitzer and Skinner[1436] reported zinc-fulvic acid stability constants ($^{log}10$) of 1.73 at pH 3.5 and 2.34 at pH 5.0. Because bonding of metals to fulvic acid involves carboxyl groups, the dependence of pH on the stability constants reflects the competition with hydrogen ions. Whenever possible, it would be preferable to eliminate the pH dependence on the metal-fulvic acid complexes in determining stability constants.

The log κ value of 5.6 at pH 7.0 for a zinc-soil-organic matter complex reported[705] indicates that most of the zinc may have been adsorbed in the form of a zinc-humic acid complex. The pH drop of the humic acids on addition of metallic cations was found to be Mn^{+2}, Ni^{+2}, Zn^{+2}, Cu^{+2}, Al^{+3}, and Fe^{+3} in descending order. Therefore, copper, aluminum, and iron may be more tightly bound than zinc to humic acid.

Zinc can be fixed through solid phase adsorption (including complexation) or the formation of insoluble precipitates. The hydrolysis (log κ_1 = 9.7) and solubility (log κ_{so} = -16.6) constants for zinc hydroxides indicate that these insoluble forms of zinc cannot be controlling the levels of soluble zinc in

soils. Most of the zinc immobilization is attributed to surface adsorption and complexing by organic matter.[711] Jensen and Lamm[793] found a high degree of correlation between zinc content and organic matter distribution in different soils. Destroying the organic matter of a surface soil allowed almost all of an extra source of zinc to be extracted with dithizone ($C_6H_5 \cdot N:N \cdot CS \cdot NH \cdot NH \cdot C_6H_5$) whereas only 50-75% could be recovered when organic matter was allowed to remain.[87] Thus, removing organic matter appears to decrease the immediate reactivity of zinc in soils.

Although the immobilization of zinc by stabilized organic matter is well established, newly formed organic substances (especially biochemical intermediates and fulvic acids) are mobile and can solubilize zinc and increase the metals availability to plants and other biologic systems. Leachates from organic soils and humic layers from forest soils often contain considerable amounts of soluble organic substances during periods of high biologic activity. Hence, zinc movement and availability can be increased greatly whenever zinc-enriched sewage sludges, animal manures, and industrial wastes are added to soils. The possibility of influencing the environmental impact of added or native zinc through proper management of soil organic matter is of considerable importance because of the variety of chemical reactions that occur between zinc and organic materials.

ZINC IN WATERS

Most modern determinations of the zinc content of sea water are in the range of 1-27 µg/l zinc, with a median at about 8 µg.[543,1291,1349,1498,1530,1625] It has been estimated that about 700,000 metric tons of zinc are transported to the sea annually.[107] More than 99.9% of the zinc reaching the sea in the dissolved form is eventually precipitated with oceanic sediments, chiefly with clay minerals, but partly with manganese oxide nodules and phosphorites. Appreciable amounts of zinc can be precipitated as sulfide in anoxic waters, such as those that exist in parts of the Black Sea.[1531]

The zinc content of fresh waters is more variable, but uncontaminated fresh waters generally contain 10 µg or less zinc/l water. The chemistry of zinc in such waters has been reviewed recently by Hem,[662] who has compared observed concentrations of zinc with those calculated from thermodynamic data. Zinc carbonate or zinc orthosilicate were assumed to be solid phases. Most surface waters appear to be unsaturated by the carbonate; the equilibrium of zinc silicate may be more important.

Recent analyses of filtered surface waters of the United States are summarized in Table 3-4. An earlier study[869] presented similar results. Zinc was detected (sensitivity 20 µg/l) in 76.5% of 1,577 samples; the maximum content reported was 1,183 µg/l, and the mean content was 64 µg/l.

TABLE 3-4

Zinc Content of Filtered Surface Waters of the U.S.[a]

Zinc Content, µg/l	No. of Samples
< 10	205
10 - 19	112
20 - 29	104
30 - 39	57
40 - 49	37
50 - 59	35
60 - 69	23
70 - 79	12
80 - 89	11
90 - 99	11
100 - 109	11
110 - 119	13
120 - 129	11
130 - 139	9
140 - 149	7
150 - 199	20
200 - 249	17
250 - 299	2
300 - 499	8
500 - 999	2
1,000 - 4,500	5
19,000	1
42,000	1
TOTAL	714

409

[a]Data from Durum et al.

High amounts of zinc in surface waters represent industrial and urban pollution, from such sources as zinc dissolved from galvanized pipes and the dumpings of plating baths. Streams that drain from areas of mining activity also may have zinc contents up to 21,000 µg/l.[424,1068] High amounts of zinc were found in waters of the South Fork of the Coeur d'Alene River (in northern Idaho) in an area where thousands of tons of ground tailings of zinc ores were exposed.[1068]

Waters of such streams tend to purify themselves by precipitating zinc with clay sediments or with hydrous iron and manganese oxides. Quantitative data are insufficient to permit an overall estimate of the proportion precipitated, but the effect of such precipitation was shown by Elderfield et al.,[424] who found 1,000-10,000 ppm zinc (average was 3,700 ppm) in sediments of the Conway River in Wales, and by Banat et al.,[69] who examined seven German rivers for zinc content, and reported highest values for the sediments of the Weser River, which contained 400-3,100 ppm zinc, with an average of 1,572 ppm. Perhac[1225] found that although the zinc content of suspended matter in two Tennessee streams was much higher than the concentration of dissolved zinc, most of the zinc was transported as dissolved material.

Zinc is picked up by water in the distribution system and household plumbing, and almost all drinking water has a detectable concentration of zinc. All but three of 2,500 samples

of water collected at consumers' taps had more than 1 µg zinc/1. The survey of 969 water systems located in nine geographic areas in the U.S. reported the average zinc content of drinking water as 194 µg/1. The highest concentration detected was 13,000 µg/1, and 0.3% of the 2,595 samples exceeded the drinking water standard limit of 5 mg/1.[1646]

When the intakes of food and water are compared, it is found that drinking water would provide 4.3% of the average food-zinc intake in the United States.

ZINC CONTENT OF COALS

Although hundreds of samples of coal have been analyzed for zinc, a meaningful average content is not easy to obtain. Nearly all the available analyses were made by spectrographic analysis of the residual ash of the coal; there is considerable danger of volatilizing part of the zinc during ashing unless special precautions are taken. Furthermore, the spectrographic method for zinc is comparatively insensitive; various workers[5,925,1823] have given their limits of detection of zinc as 50 to 200 ppm zinc in the ash. As indicated in Table 3-5, a substantial percentage of the analyses report zinc as "not found." Because all such determinations were calculated as zero, all averages given are minimal figures.

No attempt has been made to average all the data reported. Tables 3-5 and 3-6 give data based on reports in which analyses are given of a substantial number of samples, and in which the percentage of ash was given, so that results could be calculated back to the zinc content of the coal proper. Earlier data have been reviewed by Clarke and Swain[289] and by Abernethy and Gibson.[4]

TABLE 3-5

Zinc Content of Coals of the United States, by Regions

Area	No. of Samples	Average Zinc in Coal, ppm[a]	No. of Samples Reporting "Not Found"
Eastern province[b]	600	21.4	-
Appalachian region[c]	378	8.2	296
Interior province[b]	123	78.0	-
Eastern interior region[d]	475	44.0	260
Western states[b]	104	25.3	-
Western region[e]	44	28.0	-
Northern Great Plains[f]	221	59.0	199
TOTAL	1,945	AVERAGE 32.6	

[a] In these calculations, "not found" was calculated as zero; the averages are therefore all minimal figures.

[b] Derived from Abernethy et al.[5]

[c] Derived from Zubovic et al.[1824]

[d] Derived from Zubovic et al.[1823]

[e] Derived from Zubovic et al.[1822]

[f] Derived from Zubovic et al.[1825]

TABLE 3-6

Zinc Content of Coals from Major Regions of Documentation

Area	No. of Samples	Average Zinc in Coal, ppm[a]
Alabama[b,c]	137	9.7
Arkansas[d]	67	7.2
Colorado[b]	40	33.0
Illinois[b,e]	319	141.0
Illinois Basin (Ill., Ind., Ky.)[h]	82	313.0
Indiana[b,e]	123	33.0
Iowa[b,d]	25	41.0
Kentucky[b,c]	198	10.7
Ohio[b,c]	208	20.5
Oklahoma[d]	93	13.7
Pennsylvania[b]	117	22.2
Tennessee[b]	40	17.3
Utah[b]	23	76.0
Virginia[b]	51	22.7
W. Virginia[b,f]	938	32.9
East Germany[g]	494	149.0
Nova Scotia[h]	182	25.0

[a] When zinc was not found, it was calculated as zero; the averages are therefore all minimal figures.

[b] Derived from Abernethy et al.[5]

[c] Derived from Zubovic et al.[1824]

[d] Derived from Zubovic et al.[1822]

[e] Derived from Zubovic et al.[1823]

[f] Derived from Headlee and Hunter.[655]

[g] Derived from Leutwein and Rosler.[925]

[h] Derived from Ruch et al.[1382a]

The average content of zinc in U.S. coals given in Table 3-5 (32.6 ppm) [107] is of the same order of magnitude as other recent estimates: 50 ppm, 20 ppm for hard coal, [1816] and 60 ppm for brown coal. [1016] The last two estimates were based on a survey of world literature, weighed according to the total estimated reserves in each area.

The nature of the bonding of zinc in coal has been discussed by Zubovic [1821a] and Ruch et al. [1382a] There are notable areal variations in the proportion of zinc bound to the organic and to the inorganic constituents of coal, but a large proportion of the zinc is bound to inorganic constituents, and especially to the occurrence of sphalerite.

Data on zinc content of petroleum are scarce. A recent estimate for oil was 0.25 ppm, [107] and an average of 4.2 ppm zinc was given for 3 residual heating oils. [369]

CHAPTER 4

MAN-MADE SOURCES OF ZINC

MINING AND CONCENTRATING

There are over 30 mines and smelters where zinc is mined and produced in the United States. Primary areas of zinc production are mapped in Figure 4-1 and the specific locations are listed in Table 4-1.

Zinc ore, primarily sphalerite, ZnS, is mined by conventional underground mining methods, then crushed in mills, and concentrated by differential flotation. The choice of concentration techniques depends upon the chemical composition of the zinc compounds and also the other constituents (e.g., lead, copper, iron) in the ore. Losses of zinc to the atmosphere from mining, milling, and concentrating are comparatively small, but some do occur during blasting, ore handling, crushing, and wind loss from tailings. During grinding and flotation, the ore is wet and atmospheric emissions are small. It has been estimated that zinc emissions to the atmosphere from mining and milling are 0.1 kg/metric ton of zinc mined.[369] Based on the 1973 total of 435,318 metric tons for U.S. mine production of zinc, the total zinc emissions to the atmosphere from mining and milling would have been 43.5 metric tons.[1644a] However, mining and milling, including flotation, can be an important source of water-borne zinc, whenever water pumped from the mine is utilized in the concentrating process. Figure 4-2 is a flow diagram of mining and milling waste water from a lead-zinc mine in Missouri, and Table 4-2 shows the concentrations of zinc found at various stages along that path.[1796]

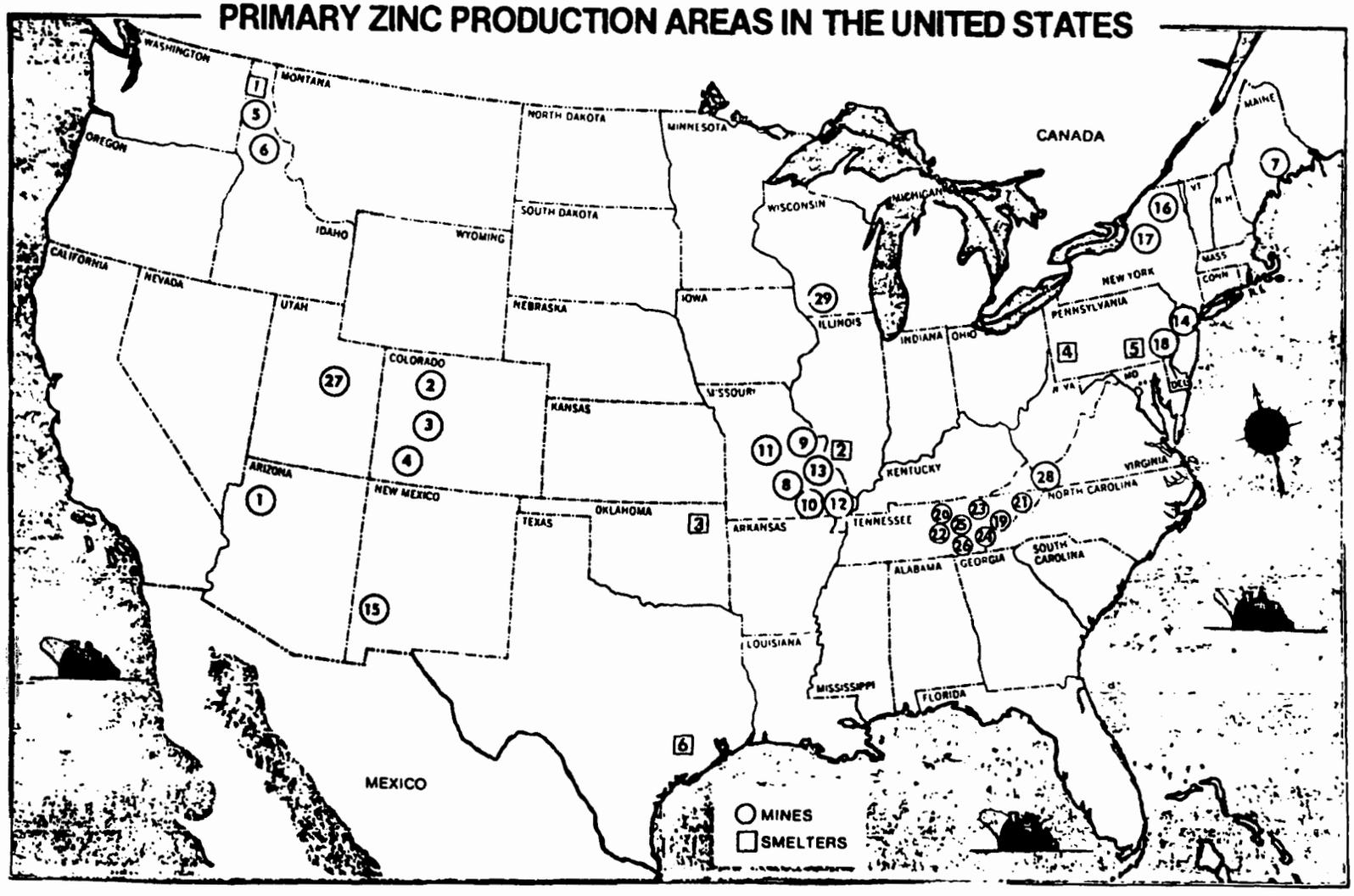


FIGURE 4-1 Primary zinc production areas in the United States. Prepared by the Zinc Institute, Inc., June 1975.

TABLE 4-1

Primary Zinc Production Areas in the United States[a]

STATE	MINES NO.	DISTRICT	COUNTY
Arizona	(1)	Bruce	Yavapai
Colorado	(2)	Eagle	Eagle
	(3)	Idarado	Ouray
	(4)	Leadville	Lake
Idaho	(5)	Bunker Hill	Shoshone
	(6)	Star Unit	Shoshone
Maine	(7)	Blue Hill	Hancock
Missouri	(8)	Brushy Creek	Reynolds
	(9)	Buick	Iron
	(10)	Fletcher	Reynolds
	(11)	Magmont	Iron
	(12)	Ozark	Reynolds
	(13)	Viburnam	Iron
New Jersey	(14)	Sterling	Sussex
New Mexico	(15)	Ground Hog	Grant
New York	(16)	Balmat	St. Lawrence
	(17)	Edwards	St. Lawrence
Pennsylvania	(18)	Friedensville	Lehigh
Tennessee	(19)	Coy	Jefferson
	(20)	Elmwood	Smith
	(21)	Idol	Hancock
	(22)	Immel	Knox
	(23)	Jefferson City	Jefferson
	(24)	New Market	Jefferson
	(25)	Young	Jefferson
	(26)	Zinc Mine Work	Jefferson
Utah	(27)	Burgin	Utah
Virginia	(28)	Austinville	Wythe
Wisconsin	(29)	Shullsburg	Lafayette

TABLE 4-1
(Continued)

SMELTERS

STATE	NO.	DISTRICT	COUNTY
Idaho	[1]	Kellogg	Shoshone
Illinois	[2]	Sauget	St. Clair
Oklahoma	[3]	Bartlesville	Washington
Pennsylvania	[4]	Monaca	Beaver
	[5]	Palmerton	Lehigh
Texas	[6]	Corpus Christi	Nueces

[a] Prepared by the Zinc Institute, Inc., June 1975.

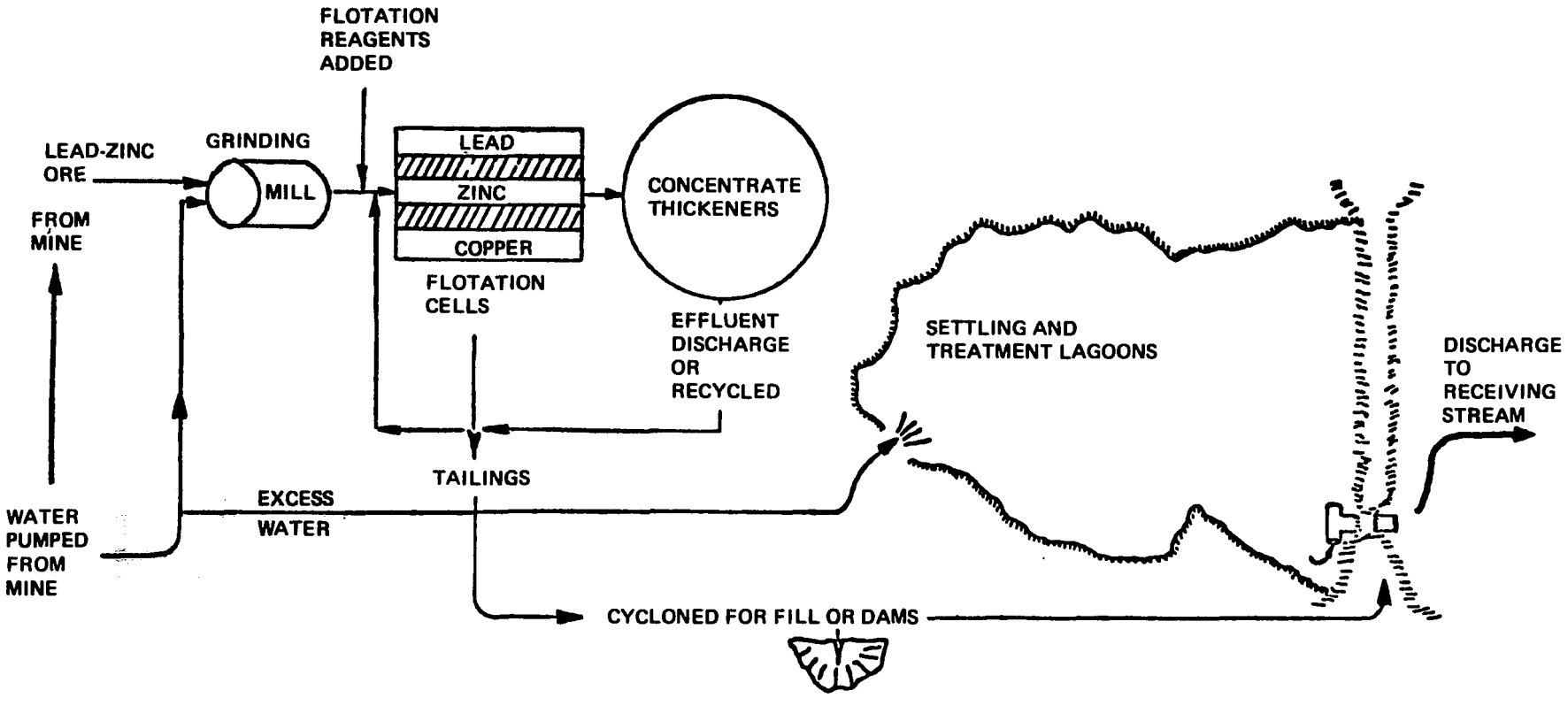


FIGURE 4-2 Flow diagram of mining and milling wastes. Reproduced from Wixson and Bolter.[1796]

TABLE 4-2

Mean Zinc Concentrations in Mined and Milled Waste Water[a]

	Locations Around Mine	Early Mining Activity zinc, ppb	Present 1971 Mining Activity zinc, ppb
Mine A (Lead, zinc, and copper ore)	Mill	11	180
	Lagoon	43	1,320[b]
	Discharge	46	1,045[b]
	Stream	24	231
Mine B (Lead and zinc ore)	Mill	139	449
	Lagoon	137	262
	Discharge	123	411
	Stream	98	328

[a]Data from Wixson and Bolter. 1796

[b]Mill discharged through tailings direct to stream during construction of new lagoons.

Wixson and Bolter[1796] hold that the differences between the final stream concentrations of zinc in the early and present mining situations were caused by reduced retention time in recent years as the lagoons filled up with tailings. However, the data in Table 4-2 indicate that the concentration of zinc in the mill water, the point furthest up the waste stream sampled, may be the determining factor of the eventual discharge concentration. It also was noted that the zinc content of the streams returned to baseline levels within a few miles of the source.

A study of the Coeur d'Alene River system in northern Idaho confirmed that nonferrous metals mining and related activities can result in high concentrations of zinc in waters that receive effluents.[1067] Zinc concentrations during low volume flow ranged from less than 0.1 mg/l for the North Fork of the Coeur d'Alene River, which has little mining activity, to 21.0 mg/l at a station in the South Fork, which has considerable mining activity. Concentrations of zinc in the South Fork were generally between 1-2 mg/l; in the main stream the concentrations of zinc were between 2-5 mg/l.[1067]

Although mining and concentrating activities are only minor sources of zinc to the atmosphere, such activities can cause significant quantities of zinc to be discharged into waterways. It should be noted that sedimentation may cause concentrations of zinc in steams receiving effluent from mining and concentrating activities to be reduced with distance downstream from the point of discharge.[1676]

SMELTING AND METALLURGIC OPERATIONS

Primary Zinc Production

There are three basic types of primary zinc smelters in the United States: horizontal-retort distillation units, vertical-retort distillation units, and electrolytic plants. Regardless of the type of smelting process zinc may be released into the air during concentrate handling, open storage, and conveying. In each of these smelting processes, the concentrated zinc ore must go through a roasting procedure to drive off sulfur dioxide, SO_2, and convert the zinc sulfide, ZnS, to zinc oxide, ZnO. Roasting may create large amounts of dust, but because that operation is enclosed, the dust may be readily collected.

The horizontal-retort distillation units are essentially batch processes in which a roasted concentrate and coke mixture are heated in a retort to approximately 1,100 C; natural gas is the usual fuel. Zinc is then reduced from the roast (zinc oxide) to zinc metal in vapor form. The zinc vapor passes into a condenser where it condenses into a liquid; it is then drained off at intervals into molds.

The vertical-retort distillation process produces highly pure zinc continuously. The basic process is essentially the same as for the horizontal-retort process, except that the charge, consisting of briquettes of zinc ore concentrate and coke, is fed into a charge column, which is a vertical extension of the retort. The charge moves down through a combustion zone where zinc vapor, carbon monoxide, and particulates are evolved. The gases pass out through the top of the column into a condenser, where the zinc vapor is condensed into zinc metal.

The electrolytic process is quite different from the other two systems. After the ore concentrate is roasted, the concentrate is leached with sulfuric acid to form a zinc sulfate solution. The liquid is then pumped into electrolytic cells where the zinc deposits on aluminum cathode. The cathodes are removed periodically and the zinc is stripped off.

The use of particulate collecting devices, primarily baghouses and electrostatic precipitators, is highly efficient (> 95%) in recovering zinc particulates. However, some emission sources are uncontrolled and little control is exercised over emissions associated with concentrate unloading, handling, and storage.[369] The particle size of zinc emissions (composed of zinc oxide and sulfur complexes) from the retort of a horizontal-retort operation were as follows: 34% of the particles were 2.5 μm in diameter, 35% were 2.5-5.0 μm, and 31% were greater than 5.0 μm.[369] Other estimates, however, have indicated smaller particle sizes. The particle size for zinc fume has been estimated at 0.01-0.3 μm in diameter and metallurgic dust and fumes at 0.001-100 μm in diameter.[317]

Few quantitative data exist on the concentration of airborne zinc near primary zinc smelters in the United States. Schrenk et al.[1441] estimated that the total pollution load of zinc oxide from a zinc smelter involved in the air pollution episode in Donora, Pennsylvania in 1948 was 11,068 kg

zinc oxide/day. Another study of two vertical-retort zinc plants estimated that the daily emissions of zinc since 1960 ranged from 6,000-9,000 kg.[198] However, particulate control systems can remove 99.5% of the particulate matter.[1238]

Based on conditions existing in 1969, the following estimates for emission factors for the three types of zinc processes were made: horizontal-retort smelting created 77 kg zinc emissions/metric tons zinc produced; vertical-retort smelting released 36 kg zinc emissions/metric tons zinc produced; and the electrolytic process produced 27 kg zinc emissions for every metric tons zinc.[369] The study also estimated that primary zinc smelting activity accounted for the discharge of 45,454 metric tons of zinc into the atmosphere in 1969.[369] Since 1969, however, five zinc smelters in the United States have ceased operations, thereby reducing the zinc-producing capacity of the United States, as well as the total emissions of zinc to the atmosphere.[1594]

Although data are not available on the concentration of zinc in the air near zinc smelters, soil analyses are available. Table 4-3, which summarizes the zinc content of soils near three smelters, shows that concentrations are much higher than the average 50-54 ppm found in normal soils. Although most of the zinc is in the top few centimeters of soil, there is appreciable downward movement, as shown by the data in Table 4-4.

Great contamination has been shown in the upper 15 cm of the soil profile near a zinc smelter.[198] The range of zinc content of soil within 1 km of the plant was 50,000-80,000 ppm zinc in the soil. Organic matter in the same area contained as much as 135,000 ppm of zinc. It has been estimated that the zinc deposition rate in this area was 1.75-5.25 g/m^3/month, or approximately 207-621 kg/ha/yr. The concentration of zinc in soil fell off sharply with distance from the plant. Background levels* of zinc were found

*The level of zinc normally found in the soil if there has been no contamination from any metallurgic operation.

TABLE 4-3

Zinc Contents of Soils near Smelters

Location	Depth of samples, cm	Distance from smelters, km	Zinc Content ppm
Swansea, W. England[c]	0 - 5	1.5	543[a]
	0 - 5	3	310[a]
	0 - 5	6	150[a]
	0 - 5	16	45[a]
Avonmouth, England[d]	1 - 5	0.32	5,000
	1 - 5	1.13	1,400
	1 - 5	4.5	450
	1 - 5	6.9	250
	1 - 5	9.5	150
	1 - 5	12.7	90
E. Helena, Montana[e]			
traverse C	2.5-10	1.61	450[b]
	2.5-10	3.22	250[b]
	2.5-10	6.44	140[b]
	2.5-10	12.87	82[b]
traverse A	2.5-10	1.61	210[b]
	2.5-10	3.22	140[b]
	2.5-10	6.44	91[b]
	2.5-10	12.87	60[b]

[a] Zinc extractable by 0.5 N acetic acid.

[b] Calculated from equation fit to data.

[c] Derived from Goodman and Roberts.[546]

[d] Derived from Burkett et al.[203]

[e] Derived from Miesch and Huffman.[1043]

TABLE 4-4

Variation with Depth of Zinc Content of Soils near Smelters

Location	Depth, cm	Zinc Content, ppm
Annaka, Japan[b]		
900 m from smelter	0 - 2	1,680
	5	1,590
	10	1,310
	20	540
	30	140
	40	80
	60	62
Poland[c]		
200 m from smelter	0 - 10	12,200
	15 - 30	1,230
	40 - 50	467
	60 - 80	57
Avonmouth, England[d]		
250 m from smelter	0 - 3	1,000[a]
	3 - 6	720[a]
	6 - 9	280[a]
	9 - 12	175[a]
	12 - 15	250[a]
	15 - 18	250[a]

[a]Zinc extractable by 2.5% acetic acid.

[b]Derived from Kobayashi.[862]

[c]Derived from Greszta and Godzik.[565]

[d]Derived from Little and Martin.[946]

25 km to the east (downwind) and 16 km to the west (upwind).[198] Therefore, airborne zinc emanating from smelters can cause significant local contamination near smelters. However, the concentrations of zinc in soil noted above are the consequences of metallurgic operations dating back to 1898. The installation of control equipment and improved control technology should mean that the area near the smelter now receives contamination at a much lower rate than in the past.

The potential waste waters from electrolytic refining are spent electrolyte solutions and slurries formed by removing impurities in the electrolyte solution by precipitating them in thickeners. According to Tallmadge, little or no zinc appears in waste water, as spent electrolyte solutions, and supernatant liquids from slurries are usually recycled for re-use in the leaching step.[1594]

A survey of lead-zinc smelting and refining operations in the United States has shown that waste waters can have zinc concentrations ranging from 0.01-25 mg/l.[606a] Tables 4-5 and 4-6 list the composition of waste waters from these processes. By comparison, a 1970 survey of surface waters in the United States indicated that most waters contained less than 0.05 mg/l; some exceeded 5.0 mg/l, and the highest value was 42 mg/l.[1116a]

Another important primary source of zinc--accounting for 68,471 metric tons of zinc in 1972[28a]--is zinc oxide from fuming furnaces. Zinc is recovered from lead blast furnace slag by heating the slag to high temperatures and blowing coal and air through it. Zinc is reduced, volatilized, reoxidized, and then collected as zinc oxide in bag filter units. Information is not available on the quantity of zinc emitted to the atmosphere during zinc fuming operations. However, the efficiency of the collecting units probably determines the magnitude of zinc arising from fuming furnaces. One survey of a fuming operation found the particulate emissions to be negligible because of the efficient collection by a baghouse unit.[1646a]

Because zinc is a constituent in other ores and raw materials, zinc dusts and fumes may be produced when other metals are refined or produced, primarily lead, copper, and steel. Athanassiadis has reported that considerable quantities of

TABLE 4-5

Composition of Waste Waters and Receiving Streams

in Lead-Zinc Metallurgic Processes for Lead Smelters and Refineries, mg/l[a]

	I[b]			II[c]		III[d]		
	Upstream	Outfall	Downstream	Intake	Receiving Stream	Intake	Neutralized Acid Plant Water	Other Waste Water
pH	7.6-8.1	7.4-8.2	7.6-8.2	7.8	7.8-11.1	7.2	5.0	8.6
Arsenic	0.11	0.15-0.46	--	--	--	--	--	--
Cadmium	0.002	0.02-1.09	0.03-0.113		0.0-0.007	--	7.7	0.5
Copper	0.13	0.13	0.13	0.34	0.0-0.015	--	0.06	4.0
Iron	--	--	--	0.08	0.0-0.03	5.0	7.4	1.3
Manganese	--	--	--	0.2	0.004-0.05	0.9	0.7	0.5
Nickel	--	--	--	--	--	--	0.2	0.06
Lead	0.03	0.07-0.157	0.03-0.05	0.14	0.0-0.01	--	0.5	11.0
Zinc	0.08-0.13	0.11-2.00	0.08-0.43	0.019	0.01-0.048	--	8.0	2.0
Sulfate ion (SO_4=)	--	--	--	6.5	9.2-23.0	126	960	200

[a] Data from Hallowell et al.[606a]

[b] Effect of waste discharge on stream: outfall was combination of processed and cooling waters. Major contributor to processed water impurities was effluent from gas conditioning operation in which fumes and dusts were moisturized by spraying them with water before entering electrostatic precipitator. Spray water was collected in sump below conditioner and discharged.

[c] Company studied contribution of waste to stream over and above background impurities by analyzing intake water and the receiving stream below the outfall. Major source of contamination in effluent was water discharged from slag granulation operations.

[d] Company operates lead smelter in conjunction with sulfuric acid (H_2SO_4) plant. It provided data on intake water neutralized acid plant water before discharge, and other waste water, which includes slag granulation and cooling water.

Composition of Waste Waters and Receiving Streams in Lead-Zinc Metallurgic Processes for Zinc Smelters and Refineries, mg/l[a]

	I[b]	II[c]	
	Effluent	Intake	Discharge
pH	7.7	5.0 - 7.0	5.2 - 6.8
Arsenic	--	- - -	- - -
Cadmium	0.39	- - -	0.9
Copper	--	- - -	0.02 - 1.35
Iron	1.0	1.0 - 1.2	1.3 - 19.7
Manganese	--	0.4 - 1.0	0.4 - 1.75
Lead	--	- - -	- - -
Zinc	8.5	5.7	18 - 25
Sulfate		136 - 179	182 - 291
Cyanide		- - -	1.3 - 1.5
Dissolved Solids	583	372 - 391	304 - 534
Suspended Solids	10	14 - 18	14 - 164

606a

[a]Data from Hallowell et al.

[b]Combined waste stream from zinc smelter includes coke plant, cooling water, gas scrubbers, spills, clean-up, etc. No sulfuric acid plant.

[c]Zinc smelter in conjunction with sulfuric acid plant. Analyses show composition of water supply and combined waste discharge.

zinc can be emitted into the air from the production of steel.[54] Average airborne zinc concentrations in four communities with steel-producing plants have been reported in Table 4-7.[1590] Concentrations during and after strikes demonstrate the impact of the industrial activity on the zinc content of the air.

Therefore, primary production of zinc and other metals may release zinc into the environment. These contributions are appreciably reduced by dust and fume control equipment and by water pollution control systems. However, even in situations under little control, the emission of zinc into the environment is likely to be confined to a limited area near the smelter.

Secondary Zinc Production

Secondary or recycled zinc is an important source of zinc in the United States. In 1972, 438,150 metric tons of recoverable zinc were produced by American mines. In addition, 83,736 metric tons of secondary zinc were produced, nearly 19% of the total U.S. zinc production.

Zinc scrap materials are composed of metallic scrap and residual scrap materials (skimmings, residues, and drosses from metallic baths). Because these materials differ considerably in zinc content and form, several different processes are employed to reclaim the metal. Scrap brass and bronze are also recovered by remelting. Bronze contains little zinc and is poured at temperatures substantially below the boiling point of bronze. Brass, however, contains 15-40% zinc and is poured at a temperature near the boiling point of brass. Therefore, major zinc emissions may occur from brass remelting operations.[369] Analysis of various brass smelter flue dust samples indicated zinc concentrations of 47.0-70.4%, when approximately 0.9-1.8 metric tons of flue dust were collected daily.[512a,1269a]

TABLE 4-7

Zinc Concentrations in Air During and After Steel Industry Strikes in Four Communities in the U.S. (1956) and Corresponding Probability Levels[a]

Community	Average Concentrations, $\mu g/m^3$ After Strike	Strike	Difference	No. of Samples	Probability[b]
Birmingham, Ala.	700	200	500	9	0.028
Donora, Pa.[c]	11,800	100	11,700	15	< 0.001
East Chicago, Ind.	1,600	300	1,300	9	0.002
Allegheny County, Pa.	1,100	100	1,000	8	0.009

1590

[a]Data from Tabor and Meeker.

[b]Probability of obtaining the observed differences by chance alone; the limit of statistical significance is p=0.05.

[c]The only community with a zinc plant (closed during the strike).

Zinc scrap materials are sorted and sometimes pretreated to drive off moisture, oil, and other organic impurities. After pretreatment, the scrap zinc-bearing material is sweated and/or distilled. In sweat processing, heat is applied to scrap materials to melt the metal. A flux may or may not be used. A molten-metal bath is formed from the metallic zinc and dissolved alloy metals. The molten metal may then be cast directly into blocks for subsequent further processing, fed directly to a distillation furnace, or cast into ingots to specifications. The distillation process is similar to the distillation systems used in primary zinc production. Either a retort or a muffle furnace is used, each equipped with condensers.[689]

The principal source of airborne zinc emissions in secondary zinc processing occurs from vaporization of the metal in melting and pouring, and through escape of zinc fume from the sweating and distillation processes. It has been estimated[369] that zinc emissions to the atmosphere vary from 5 g-62 kg/metric tons of products, averaging 9 kg/t. The total zinc emissions to the atmosphere came to 3,455 metric tons in 1969.[369] Although the amount of secondary zinc seems small when compared to emissions from primary zinc production (45,455 metric tons in 1969), as secondary zinc evolves as a more important source of the metal, its contribution to total zinc emissions will increase.

Zinc Oxide Production

Zinc oxide is used extensively in industry. In 1974, 228,356 metric tons of zinc oxide were produced in the U.S. and used for many purposes, the most important being in the production of rubber.[1354a] Zinc oxide may be produced chemically, or by direct or indirect pyrometallurgic means. As mentioned, in the direct (American) process, zinc vapor is produced from ore or scrap and

then oxidized to form zinc oxide. The indirect (French) process uses zinc metal, which first is vaporized and then oxidized to form zinc oxide. One survey has indicated that zinc emissions to the atmosphere during the production of zinc oxide in 1969 ranged from 9-85 kg zinc/metric ton of product. It was estimated that in 1969 emissions from zinc oxide production totalled 7,330 metric tons.[369]

Manufacturing and Fabricating

The major end uses of slab or metallic zinc are galvanizing, manufacturing brass products, and die-casting alloys. During galvanizing, zinc is primarily released into the atmosphere when ammonium chloride flux is added or when the flux layer over the hot zinc bath is disturbed.

Brass is a copper alloy containing up to 40% zinc. Bronze contains a much smaller percentage of zinc. During processing of both products, the metals are melted together and poured at elevated temperatures (649-1,316 C). Zinc fume is released to the atmosphere during heating and pouring. Similarly, when zinc die-castings are produced, the zinc alloys are melted and poured into dies and then cast into the desired forms.[369] Table 4-8 lists the amounts and sources of zinc emissions into the atmosphere from manufacturing and fabricating.

Industrial operations using zinc contribute to the zinc content of streams into which waste water flows, of course, but few quantitative data exist from individual sources.

ZINC IN THE COMMUNITY ENVIRONMENT

Available data on air levels of zinc in urban communities without mines or smelters indicate a general decline in airborne zinc from 1954 through 1964.[54] Lee and Lehmden[914a] report concentrations of zinc particulates in urban areas

TABLE 4-8

Airborne Emissions from End Uses of Zinc[a]

Use	Major Source of Emission	Emissions, metric ton/yr (1969)
Zinc-based alloys	Melting	2,727
Zinc-coating	Molten zinc baths	864
Brass and bronze	Melting	164
Rubber tires	Abrasion	7,636
Photocopying	Waste disposal (burning)	1,364
Paint	Production	9
Zinc sulfate	Production	27
Miscellaneous		1,000

[a]Adapted from Davis.[369]

throughout the U.S. ranging from 0.1-1.7 $\mu g/m^3$, and data from the National Air Sampling Network have recorded annual average airborne zinc concentrations throughout the U.S. that are generally less than 1 $\mu g/m^3$, as shown by Table 4-9.[1645a]

Motor vehicles, fuel oil and coal combustion, incineration, soil erosion, and industrial, commercial, and construction activities contribute to zinc in urban atmospheres. Estimates have been made of the amounts of zinc released into the urban atmosphere by these activities and products. For example, tire wear and leakage and combustion of fuels and lubricants containing zinc additives account for most of the zinc released by motor vehicles. Rubber tires contain 1.5% zinc by weight;[1172a] according to Davis,[369] zinc wears off rubber tires at a rate of 45 g zinc/tire/yr, or 1.2 kg zinc/million km. Fuel and lubricating oils contain 30-1,500 ppm zinc;[1172a] the zinc arising from motor vehicle emissions is preferentially attached to small (submicron) particles in amounts of 0.1-10 ppm.[914a]

The amount of zinc contributed from motor vehicles appears to vary from city to city. Gordon, in College Park, Md., and Friedlander, in Los Angeles, reported that the fraction of zinc in the atmosphere accounted for by motor vehicles was 23% and 22%, respectively.[1172a] However, Creason *et al.*[335] found that neither site differences, distances, nor depth gradient effects were significant for dustfall zinc when comparing samples representative of various industrial and urban patterns collected from roadside sites in Cincinnati. Ondov *et al.*[1172a] compared zinc:lead ratios for urban area roads and a tunnel, which provided a system for accurate measurement of background air. Samples from the tunnel gave a zinc:lead ratio of 1:100, whereas the urban areas had a zinc:lead ratio of 1.10:0.25; hence, motor vehicles account for only a small percentage of the zinc observed in city air. Moreover, they are not the major source of zinc on suspended particles in urban atmospheres.

TABLE 4-9

Urban Air Samples and Yearly Zinc Averages, $\mu g/m^3$ [a]

Birmingham, Alabama	1.09	Baltimore, Maryland	.34
Paradise Valley, Arizona	.05	Brockton, Massachusetts	.11
Texarkana, Arkansas	<.01	Kalamazoo, Michigan	.04
Bakersfield, California	<.01	Moorhead, Minnesota	<.01
Bridgeport, Connecticut	1.60	Lincoln, Nebraska	.50
Atlanta, Georgia	.52	Bridgeton, New Jersey	.08
Boise, Idaho	<.01	Albuquerque, New Mexico	<.01
Moline, Illinois	<.01	Akron, Ohio	.48
Beverly Shores, Indiana	.20	Altoona, Pennsylvania	.27
Dubuque, Iowa	<.01	Baymon, Puerto Rico	<.01
Ashland, Kentucky	.54	Portsmouth, Virginia	.08

[a]Adapted from the National Air Sampling Network.[1645a]

Fuel oil is used in homes for heating and by industries, electric utility companies, railroads, oil companies, and the military. Based on an average zinc content of 4.17 ppm for fuel oil, Davis[369] has calculated a zinc emission factor for fuel oil of 0.64 kg zinc/1,000 barrels of oil burned. In the U.S. in 1969, combustion of fuel oil accounted for 410 metric tons of zinc emitted to the atmosphere, or approximately 0.28% of total zinc emissions. The probable forms of the zinc particles in the flue gas are zinc oxide and zinc sulfate.[860a]

Davis[369] also provides estimates for zinc emissions contributed by burning coal. The average zinc content of various U.S. coals is 54.6 ppm (see Chapter 3). Based on U.S. figures for 1969, the calculated zinc emission factor for burning coal is 8.5 kg zinc/1,000 metric tons of coal burned. This means that an estimated 3,922 metric tons of zinc were emitted, that is, 2.7% of the total zinc emissions from milling and nonmining sources. Most of the zinc is discharged with the airborne fly ash.[1120a]

Small but significant amounts of zinc are released during incineration. Davis[369] calculated that 25,435 metric tons of zinc, or 17.5% of total zinc emissions, were emitted into the atmosphere in the U.S. in 1969 from incinerations. This estimate is based on an average zinc content of about 2,400 ppm[291a] in sewage, sludge, and 23,842 metric tons of refuse and garbage that were ultimately incinerated.

Much of the zinc particulate material released into urban atmospheres from all of these sources settles on pavements as "fallout" and is washed into natural and man-made drainage systems as rainfall runoff. A study of runoff from street surfaces following a moderate-to-heavy storm in seven cities revealed zinc values ranging from a low of 0.03 kg/curb mi to a high

of 0.95 kg/curb mi, with an average of 0.34 kg/curb mi (see Table 4-10).[1414a] The zinc was of no particular size or particle range, and it was one of the metals present in the greatest amount in the runoff, regardless of land use category (see Table 4-11). Zinc loading intensities were heaviest in industrial areas and lightest in commercial areas (see Figure 4-3). However, such distinctions disappear when the contribution of zinc is considered in terms of percent by weight of total solids, that is, pounds (grams) of metal per 100 pounds (kilograms) of dry solids, as shown in Figure 4-4.

Drinking water samples from throughout the United States indicate that despite the contribution of airborne zinc to water systems as fallout or in precipitation, the drinking water standard of 5 mg/l was exceeded on the average in only one of 969 water systems tested. Of a total of 2,595 distribution samples, the 5 mg/l standard was exceeded in 8 cases, or 0.3%. The maximum zinc concentration found was 13 ppm.[1646]

Available data indicate that significant zinc contamination of the environment in specific limited areas near point sources exists. However, there do not seem to be major mobile sources of zinc nor does zinc appear to be an increasing environmental contaminant.

ZINC USES AND MARKETS

Zinc consumption in the United States during the 1960's and on into the 1970's has averaged over 1,183,000 metric tons/yr, making zinc the fourth most used metal after steel, aluminum, and copper. The consumption rate is high because of the wide availability and advantageous properties of zinc.

Consumption of zinc is sensitive to economic cycles, particularly because the metal is mostly used in automobile production (for die-casting,

TABLE 4-10

Concentrations of Zinc in Runoffs from Street Surfaces[a]

City	Zinc Loading Intensity, kg/curb mi
San Jose - I	0.63
San Jose - II[b]	0.13
Phoenix	0.16
Milwaukee	0.95
Baltimore	0.59
Atlanta	0.05
Tulsa	0.03
Seattle	0.17
Average	0.34

1414a

[a]Derived from Sartor and Boyd.

[b]San Jose was tested twice, once in December 1970 and again in June 1971; other cities were tested in the summer of 1971.

TABLE 4-11

Distribution of Heavy Metals by Land Use Category from Street Surface Contaminants, % by Weight[a]

Metal	Residential[b]	Industrial	Commercial	Total
Chromium	5	8	5	7
Copper	10	14	20	11
Zinc	38	44	24	40
Nickel	1	5	3	3
Mercury	10	4	20	4
Lead	36	25	28	35
Cadmium	--	--	--	--
	100%	100%	100%	100%

[a] 1414a
Derived from Sartor and Boyd.

[b] The figure reported here as "residential" was computed by combining all of the observed data for the four residential land use categories sampled in each city. "Industrial" and "commercial" figures were computed similarly.

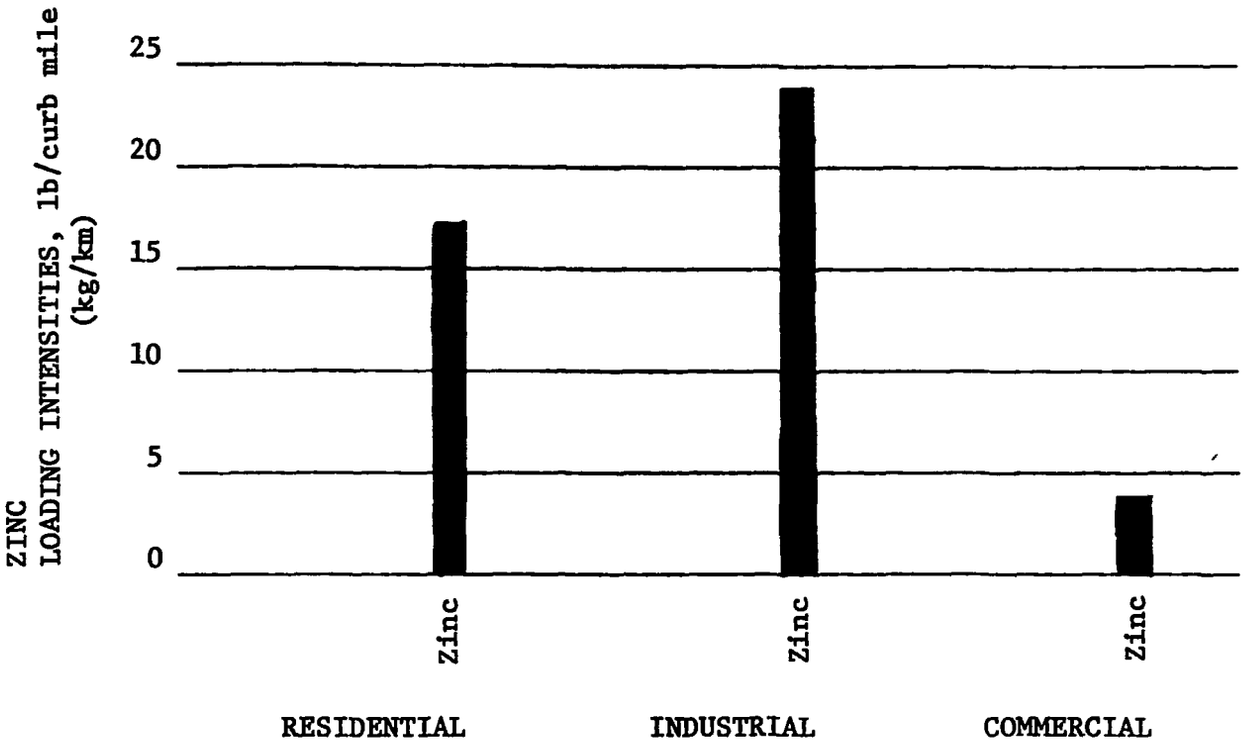


FIGURE 4-3 Zinc loading intensities on street surfaces: variations with land use. All seven cities are considered together. The metric equivalent for residential areas is about 12.24 kg/km; for industrial areas, it is about 17.28 kg/km; and for commercial areas, the figure is about 2.16 kg/km. Reproduced from Sartor and Boyd.[1414a]

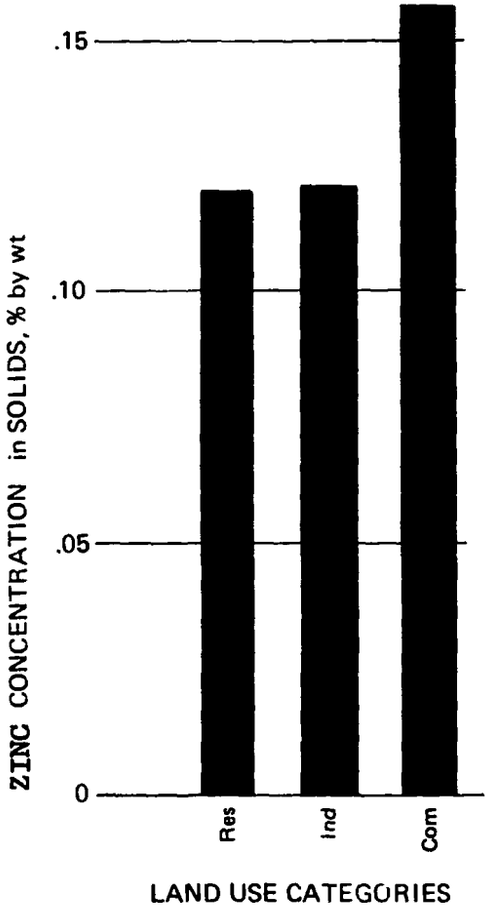


FIGURE 4-4 Varying zinc concentrations according to land use. All seven cities are considered together. Reproduced from Sartor and Boyd.[1414a]

galvanizing, and oxide for rubber tires) and in industrial and residential construction activity (for galvanizing, brass, die-casting for appliances, and oxide for paint pigments). This sensitivity can be observed by noting the recent change in the consumption pattern for zinc in the United States. Since the 1960's, die-casting alloys accounted for the largest consumption of zinc, galvanizing has been second, and brass a distant third.[1354a] These patterns of consumption are charted in Figure 4-5.

In 1974, however, the U.S. consumption of zinc for die-casting alloys dropped to a level below that for galvanizing. Because of anticipated growth in the galvanizing markets and a probable lower growth rate in zinc die-casting, consumption of zinc for die-casting is likely to remain at a lower level (personal communication, T. F. Shaffer, Zinc Institute, Inc.). Consumption patterns vary throughout the world, but in general, galvanizing is the leading market, especially in Japan and Australia, where zinc consumed for galvanizing surpasses all other zinc markets combined. In Europe, galvanizing is also the leading zinc market in all countries except the United Kingdom, where brass is the largest single consumer of zinc.

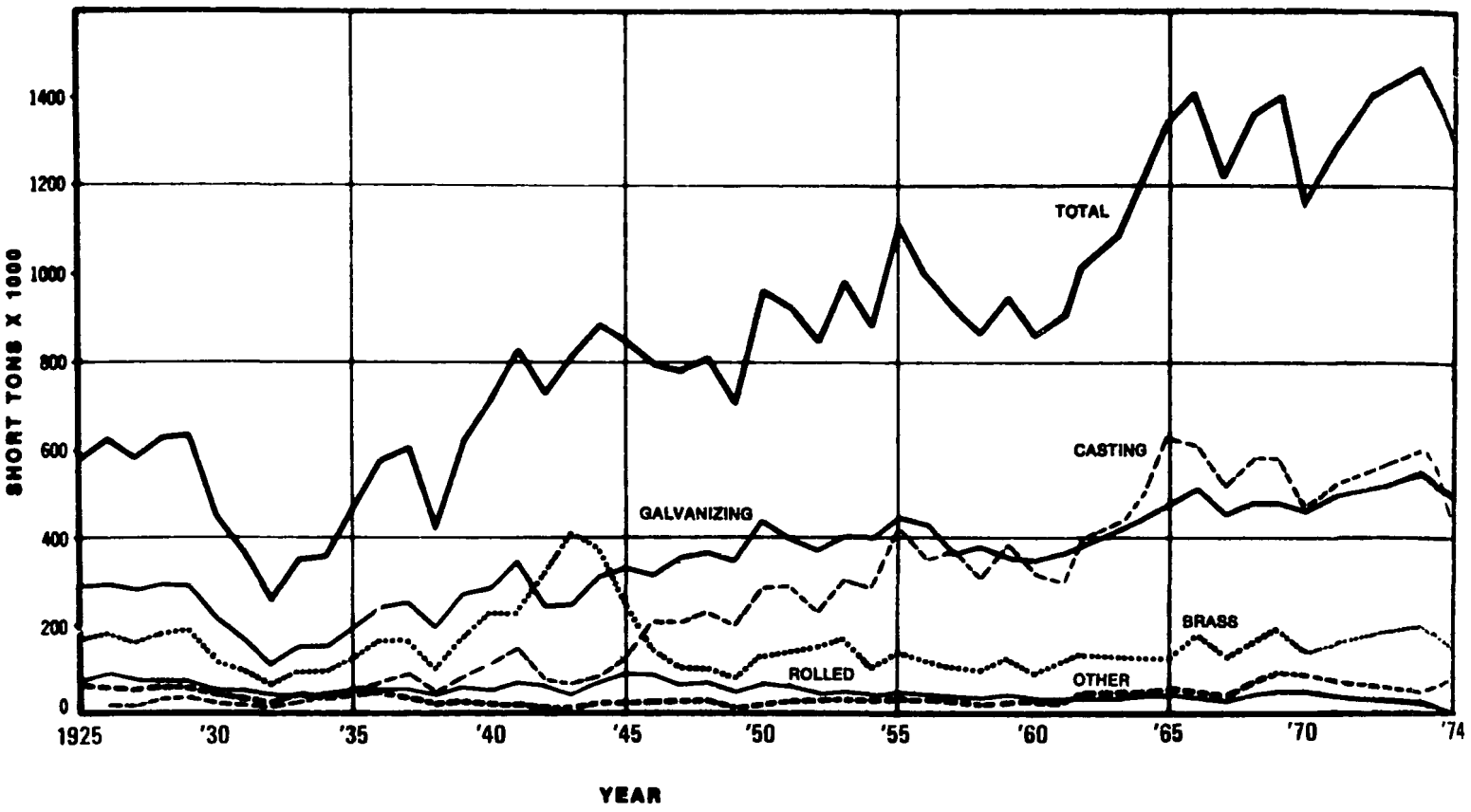


FIGURE 4-5 Consumption of slab zinc in the United States. Reproduced from the annual review of the U.S. zinc industry.[1354a]

Zinc Die-Casting

For many years, zinc alloys have been the most widely used metal in die-casting, because of their combination of favorable properties, adaptability to the process, and low cost. The low melting point of zinc also preserves the life of the dies longer.

It has been estimated that over 455,000 metric tons of zinc were used in 1974 by die-casters in the United States.[1354a] The metal is used in major appliances and automobiles for functional parts like pumps, impellers, carburetors, and housings. It is also employed for decorative door handles, trim, frames, and control panels. Toys, hardware, plumbing fixtures, and industrial components are other major markets for zinc die castings.

The development of ILZRO 12,* a general foundry zinc alloy, in the late 1960's has opened new casting markets for zinc. Components previously cast in iron, copper-base, or some aluminum alloys are now being produced with this zinc alloy.

New die-casting techniques developed in the early 1970's have made it possible for conventional zinc castings to be made with cross sections only about half as thick as those made in previous years. Thus, material costs and weight of the product are reduced, yet its tensile, fatigue and impact strengths are retained or even improved.

*Mention of this product does not imply any endorsement or recommendation by the National Academy of Sciences or the National Research Council.

Wrought Zinc

Depending on the ultimate product requirements, numerous compositions and alloys are used for wrought zinc applications. Various alloying metals are used to improve various properties (such as stiffness and creep resistance*) for specific applications.

The conventional grades of slab or ingot zinc are rolled into sheet, strip, ribbon, foil, plates, or rod according to their uses. For instance, dry cell battery cases, are made from wrought zinc rolled into a strip. The zinc also may be continuous cast+ into rod or bar. Because of its properties, rolled zinc is easily workable into various shapes and forms by common fabricating methods, such as stamping, forming, and spinning. It may be polished, plated, painted, or left to weather naturally.

Major markets for wrought zinc include the electrical and electronics industries, which make products such as brackets, shielding, capacitor cans, and heat sinks; general manufacturing, for products like drawer knobs, bezels, and golf club sole plates; the jewelry industry for medallions, jewelry, and buckles; and construction, for flashing, trim, and decorative panels.

Galvanizing

One of the oldest functions of zinc has been as a protective coating on steel to prevent rusting and its ultimate destruction. It is estimated that over 450,000 metric tons of zinc were consumed in the U.S. in 1974 for coating of

*Creep resistance is the ability of metal to resist elongation over long periods of time under sustained loading stress. See also Chapter 2.

+Continuous casting is a process for converting molten zinc directly into rods without interruption by casting the metal between two counter-rotating steel wheels with V-slots on their faces.

Continued improvements in the galvanizing process and related areas have increased consumption totals markedly for conventional applications. In the late 1930's, for example, the introduction of the continuous hot-dip galvanizing line led to wide installation of such lines around the world, turning out millions of tons of galvanized sheet and strip. But the traditional method of dipping a steel product into a bath of molten zinc is still very much in use, particularly for prefabricated and very large parts. Over the years, galvanizing has proved effective in protecting such steel products as bridges, railroads, automotive vehicles, ships, wire, cable, pipe, hardware, containers, trash cans, and roofing.

Although hot-dip galvanizing is the most popular method for protecting steel against corrosion, metallic zinc can be applied to steel in several other ways: by electrodeposition, metallizing or spraying of molten metal, sherardizing, and painting.

The all-galvanized bridge became a reality in the late 1960's when it was demonstrated that galvanized bolted connections were technically and economically feasible.[31a] The use of galvanized steel reinforcement for concrete also became widely accepted then. Galvanized steel reinforcements are integral parts of major governmental and institutional buildings, office buildings, bridges and roads, chemical plants, storage tanks, and other buildings where prevention of concrete spalling or rust staining from black steel reinforcement is necessary.

Zinc-rich paint has become one of the fastest growing new markets for zinc in the past few years. Such paints are most often used to supplement galvanized steel for automotive underbody protection against corrosion, for

bridges and other structures, for ship hulls, and for various buildings. Zinc-rich paint provides sacrificial galvanic protection of steel similar to that of galvanized coatings and is recommended whenever economics or size and shape of products make conventional galvanizing impractical.

Large-scale applied research in the early 1960's on whether zinc anode systems should be used for marine tankers showed zinc to be superior to magnesium anodes in performance and economy. That zinc does not spark was a significant advantage, and now zinc anodes are the preferred sacrifical anode for protection against corrosion in this application. Related research led to a later development of zinc anode systems for galvanized hot water storage and glass-lined tanks, which provided superior service life at an economic price. Protective zinc anodes also were developed for crab pots.[198a]

Zinc Chemicals

Zinc and its compounds are used more and more in the chemical-metallurgic, ceramic, fertilizer, paint, paper, plastics, rubber, textile, and electronics industries. As mentioned, zinc oxide is the most important zinc chemical; it is employed primarily as a reinforcing pigment in rubber, where it provides good heat conductivity and resistance to aging by sunlight. Zinc oxide is a necessary substance for the copying industry, where it is used as a thin electrostatic coating material. Although only a micron-thick layer of zinc is applied when used, the zinc oxide consumed by the copying industry has been estimated to be around 33,000 metric tons in 1972.[1016a]

Recently, zinc has become a supplement in plant and animal feed. Trace amounts of zinc are essential for the growth and development of plants and animals, and this essentiality will be discussed in Chapters 5, 6, and 8.

WASTE DISPOSAL

Tailings

Tailings are the gangue and other refuse material left over from the washing, concentration, or treatment of ground ore. Tailings from the processing of zinc and lead ores may include considerable amounts of zinc, as well as sulfates, chlorides, copper, iron, lead, cadmium, arsenic, and selenium. These tailings are dissolved or suspended in process waste waters which have come into direct contact with raw materials and intermediate or final products or by-products. Table 4-12 shows some of the concentrations of metals contained in waste water discharge streams.[1643]

Most major waste water streams from processing plants contain sulfuric acid as well as trace metals. Addition of a lime slurry to the water raises the pH from around 2 to between 10 and 11.5, causing precipitation of several of the metals as hydroxides. The water is channeled through an enclosed system to a settling pond where the suspended solids settle; absorption of carbon dioxide from the atmosphere leads to formation of carbonates, gradually reducing the pH of the pond. The effluent which evolves contains less than 10 mg/l of total suspended solids.[1643] Table 4-13 shows typical concentrations of various constituents from a lime and settle treatment plant.[1643]

Waste water is often cycloned before liming and settling. As the water passes through cyclone pumps, coarse sand is separated from finer material. The coarse sand is used to build the retaining walls of the disposal dams; the fine material is dumped into the lagoon with the mine water or other waste stream water and left to settle. Waste water from flotation concentrators, thickeners, and zinc scavenging circuits is treated by cycloning and then allowed to settle. Cooling water from metal casting operations is usually just settled, whereas acid-plant blowdown can be treated in several ways:

TABLE 4-12

Gross Concentrations in Discharge Streams[a]

Waste Material	Concentration[b]
Dissolved solids	455-4,485 mg/l
Suspended solids	25-249 mg/l
Sulfates	175-2,221 mg/l
Chlorides	60-620 mg/l
Arsenic	0.1-0.68 mg/l
Cadmium	0.02-2.4 mg/l
Copper	0.01-0.34 mg/l
Iron	0.02-1.93 mg/l
Lead	0.02-1.35 mg/l
Selenium	0.007-1.8 mg/l
Zinc	5-243 mg/l

[a]Data from the U.S. Environmental Protection Agency.[1643]

[b]Data summarized from six smelting/refining plants.

TABLE 4-13

Effluent Concentrations from Lime and Settle Treatment of Mixed Wastes[a]

Constituent	Concentrations, mg/l[b]		
	Minimum	Maximum	Average
pH	9.3	10.8	9.6
Total solids	1,430	4,050	-
Sulfur	250	730	650
Chloride	140	490	480
Cadmium	0.03	0.7	0.3
Lead	0.5	1.8	0.7
Selenium	0.8	5.0	-
Zinc	1.0	8.8	2.0

[a] Data from the U.S. Environmental Protection Agency.[1643]

[b] Except for pH.

by a combination of liming and settling, by mixing with other waste streams, settling, and liming, or by mixing and settling. The Akita zinc smelter in Japan, designed with zero pollution emission as a primary goal, treats its waste waters by using a two-stage neutralization with ground limestone, $CaCO_3$, and calcium hydroxide, $Ca(OH)_2$, followed by settling. The final discharge water from such treatment has a zinc content of 0.5 ppm or less.[1019] Table 4-14 compares the efficiencies of various methods which remove zinc from effluents.[933]

The clarified water that remains in tailings ponds after settling occurs is left to evaporate, to percolate through the bed of the pond and underlying soil, or to be diluted by rain and snow before being decanted and released to the natural watershed. Or, it will be discharged directly to the watershed if the concentrations of trace contaminants are low enough.

Few specific data are available on the concentrations of zinc contributed to ground water from water percolating through the beds of tailings ponds. Concentrations of zinc in the natural waters of zinc mining areas have been found as high as 50 mg/l, although in most ground and surface fresh water, zinc is present only in trace amounts.[1434] A median value of 20 µg/l has been reported in the surface waters of the United States.[662]

Studies of the translocation of zinc in soils demonstrated the relative immobility of zinc added to sandy soils, even at pH 4.[890] Similarly, Wheatland and Borne, applying river water to soils of the Bunter Sandstone area of England, found the concentration of zinc in the percolate to be considerably less than the concentration in the water applied (see Table 4-15).[1762]

TABLE 4-14

Treatment Processes for Removal of Zinc[a]

Treatment Processes	Possible Effluent Concentration after Treatment, mg/l
CP[c] (with lime) + S[d]	0.5-2.5
CP[c] (with lime) + S[d] + F[e]	0.1-0.3
CP[c] (with lime) + S[d] + RC[f] + F[e] + AC[g]	0.4

[a] Data from Lin and Lawson.[933]

[b] Treatment efficiencies vary according to initial concentrations of raw wastes.

[c] Chemical precipitation.

[d] Sedimentation.

[e] Filtration

[f] Recarbonation.

[g] Activated carbon adsorption.

TABLE 4-15

Mineral Analyses of River Water and Percolate Applied to the Soakage Area[a]

Mineral, ppm	Water Applied 9/30/57	Water Applied 1/31/58	Percolate 9/30/57	Percolate 1/31/58
Total hardness (as $CaCO_3$)	262.0	--	268.0	--
Total dissolved solids	456.0	--	457.0	--
Calcium	76.0	--	80.0	--
Magnesium	18.8	--	18.0	--
Sulfate	88.0	94.0	89.0	90.0
Chloride	49.0	49.0	49.0	49.0
Iron	0.5	0.3	0.2	0.24
Nickel	0.16	0.11	0.03	0.04
Total chromium	0.04	0.015	0.015	0.017
Manganese	0.25	0.01	nil	nil
Zinc	1.00	0.25	0.06	0.02
Lead	0.025	0.01	0.012	0.008
Copper	0.035	0.02	0.01	0.02

[a] From Wheatland and Borne.[1762]

Zinc was mobilized in soil by continuous flooding in the presence of fermenting organic material, but immobilized when the soil was reoxidized.[918] Lehman and Wilson observed similar results, using suburban residential sewage effluent filtered through sandy soil material. When lysimeter columns were intermittently treated with effluent, less translocation of trace metals occurred than when the columns were treated by continuous flooding. The tests performed on both field plots and lysimeter columns proved that:

- zinc was present in approximately the same amounts in both studies; concentrations of zinc at the 15- and 30- cm depths of the field site were nearly the same as the concentrations at comparable depths of the continuously flooded lysimeter columns;
- in the lysimeter study, zinc was removed from the applied effluent at or near the soil surface, and only minor amounts of zinc were detected in the filtrates; and
- some zinc was translocated when filtration rates were high, as during the first few weeks of irrigation and when the soil was saturated.[918]

Sewage

Zinc is one of the trace elements found in highest concentration in municipal waste waters. Depending upon the treatment process utilized, the zinc in waste water will be separated into effluent and sludge. On a concentration basis, the sewage sludge is usually enriched, whereas the effluent is lower in zinc than the incoming waste water. Even though effluents may be relatively low in zinc content, the great amounts of effluent which are discharged at larger sewage treatment plants can be responsible for discharging significant portions of zinc to receiving streams, irrigation sites or other effluent-receiving systems. The zinc concentration permitted in industrial effluents is regulated by discharge limits, so the metal is removed from contaminated industrial waste waters and often forms industrial sludges which are highly contaminated by zinc as well as other metals. These sludges are usually disposed of by landfill, but some are incinerated.

Pound and Crites[1268a] have listed effluent concentrations of zinc measured from differing stages and types of sewage treatment in California. The values given for primary effluent were 0.83 ppm zinc, trickling filter effluent, 0.16 ppm zinc, activated sludge effluent, 0.32 ppm zinc, and pond effluent, 0.39 ppm zinc. These data are based on limited samples, but Blakeslee[123a] has presented data on effluents ranging from 0.01 to 4.7 ppm zinc in a survey of 58 sewage treatment plants in Michigan. However, most effluent values for zinc are well below 1.0 ppm, and a value of less than 0.5 ppm zinc has been cited as typical.[1737a] When sanitary and storm sewers are combined, the addition of zinc to sewage by fallout of zinc particulate material onto urban pavements is significant (see Figure 4-3 and Table 4-9). Therefore, many loading factors of zinc must be considered, and the zinc concentration in the effluent from a single treatment plant may vary greatly, depending on water use and amount of zinc received.

If all sewage were treated to meet secondary treatment requirements set by the EPA, about 8.1 million dry metric tons of digested sludge would be produced in the U.S. each year; the current amount is about 4.5 million dry metric tons (personal communication, R. Bastion). Because of the variation in wastes arriving at the sewage treatment plants, the sludges range widely in zinc concentrations. Many municipalities have industries that produce waste water high in zinc and more than 98% of it is segregated into sewage sludge. Sludge zinc concentration increases during anaerobic sludge digestion, because unstable organic components are lost and zinc forms an increasing percentage of the remaining material. Therefore, the zinc is effectively concentrated on a dry-weight basis.

Chaney[256] has found zinc concentrations in digested sewage sludge ranging from 500 to 50,000 ppm. Dean and Smith[371a] have shown that the distribution of zinc values in sludges tend to be logarithmically normal: some extremely high values are traceable to industrial effluents. A geometric mean gleaned from over 100 studies was 2,420 ppm zinc, and 80 additional samples from sewage treatment plants in the U.S. yielded a geometric mean of 6,380 ppm zinc.[371a] Berrow and Webber[106] have described the composition of British sludges and the range of values for the 42 sludges sampled was 700 to 49,000 ppm zinc, with a mean value of 4,100 and a median value of 3,000 ppm. By comparison, normal values for zinc in soils range from 10 to 300 ppm with a typical level of 80 ppm.[283] Swedish workers have reported a mean value of 2,800 ppm zinc in 1968 and 1969 and 2,500 ppm in 1970 from routine samplings of sludges at 57 treatment plants.[1158] The five sludges highest in zinc in Sweden ranged from 5,700 to 17,200 ppm zinc.[1158] Blakeslee[123a] reported zinc values for sludges from 58 municipal sewage treatment plants in Michigan. They ranged from 72 to 16,400 ppm, with

a median value of 3,200 ppm zinc. Recently, Page[1191a] and Peterson et al.[1236a] also have summarized published data on the metal content of sludges.

Depending upon the disposal or utilization of the sludge material, the presence of zinc in sewage sludge may or may not be a potential problem. Zinc contamination should cause no problems in well-designed landfill sites where there is no opportunity for leaching into surface waters or groundwater supplies. As mentioned, significant amounts of zinc may be released to the atmosphere when sludge is incinerated. Sludge which will be applied to agricultural or other lands that support plant growth may be the most dangerous problem. Indeed, zinc is such a common contaminant of sewage sludges that Chumbley[283] expressed the permissible levels of toxic metals in sewage used on agricultural land in terms of a zinc equivalent.

Solid Wastes

In addition to the effluents discharged to tailing ponds, zinc-bearing converter dust and retort waste are produced as solid wastes during smelting of zinc and lead ores. These wastes used to be dumped, but now the zinc is recovered.[836] Converter dust is a mixture of zinc, lead, iron, and copper oxides. Leaching with a sulfuric acid solution removes the copper; addition of a bleaching powder precipitates the iron. The remaining zinc sulfate reverts to zinc through electrolysis. Retort residues, consisting of unconsumed coal, coke, and unreduced ore from incomplete distillations, may contain 5-15% zinc. Because the composition varies so greatly, each residue must be separately analyzed to determine the best method of treatment.

Many foundry and metallurgic operations produce wastes that can be treated to recover zinc. Skimmings from galvanizing operations may contain 20% metallic zinc and 35% zinc chloride; the zinc drosses that form at the bottom of galvanizing vats may yield intermediate grades of zinc; crude zinc oxide can be recovered from the leaching vats in which skimmings from galvanizing are treated. Zinc ashes and flue dust from foundries and smelters can be mixed with zinc ore and be distilled into slab zinc, or they can be used to make oxides and pigments; die-cast drosses and turnings may contain 40-70% recoverable zinc.[944] The reclaimed zinc products are used for galvanizing vat charges, producing rubber, making paint pigments and ceramic glazes, and for rolled zinc.

Trace amounts of unrecoverable zinc will still remain in waste material even after reprocessing. This material frequently is deposited in municipal sewer systems and treated with the rest of the sewage. If the sludge from the sewage treatment is dried and incinerated, trace quantities of zinc may be released into the atmosphere. Zinc oxide is also a potential effluent from the incineration of vinyl products in which the zinc was used as a heat stabilizer. Data on the concentration of zinc emitted from one stack of a twin-stack municipal incinerator are set forth in Tables 4-16 and 4-17.[1813]

TABLE 4-16

Metals Analysis of Samples Extracted from Various Points of One Stack of a Two-Stack Municipal Incinerator[a]

	Metal Concentrations							
	Cadmium		Lead		Zinc		Copper	
	µg	µg/g	µg	µg/g	µg	µg/g	µg	µg/g
Probe backwash, suspended solids, 0.0192 g	0.845	44	124.9	6,507	78.2	4,077	8.18	426
dissolved solids, filtrate	0	0	0	0	--	0.04	0	0
Glass fiber filter, 0.2097 g	401	1,912	16,288	77,677	1.1×10^5	556,340	319	1,519
Impinger contents 345.5 ml	6.91	0.02	0	0	--	0.47		0.04
TOTAL	408.8		16,413		1.1×10^5		327.2	

[a]Data from Yost et al.[1813]

TABLE 4-17

Metals Emission Rate from One Stack of a Two-Stack Municipal Incinerator[a]

Metal	Mass Emission Rate, g/sec[b]
Cadmium	0.017
Lead	0.683
Zinc	4.580
Copper	0.014

[a] Data from Yost et al.[1813]

[b] $\mu g/sec = \mu g/m^3 \times m^3/sec$

Nuclear Reactors and Zinc-65

A major component of any nuclear reactor is the coolant material, which is essential in removing the heat released by the fission process. Water is a popular coolant because it can also serve as a moderator to slow down neutrons. Although the water is purified before circulating through the reactor, small amounts of trace elements dissolved in the water are not removed. Also, corrosion products from reactor construction materials and feedwater systems are present. These impurities concentrate on the fuel cladding surfaces where they are bombarded by the intense neutron flux in the reactor and become radioactive. Zinc-65 is one of the radioactive contaminants formed in reactor coolant water in this way, particularly in those reactors in which admiralty brass is used in the feedwater system.[232a] Zinc from aluminum alloys also provides the target material. The nuclear reactions involved are copper-63 ($^{2}H,\gamma$), zinc-65 and zinc-64 (n,γ), zinc-65.

The radioactive material is subsequently removed from the coolant water by ion exchange. Since it is generally classed as low-level, that is, it contains 10^{-4} μCi/ml or less of activity,[1645] this material may be contained for a brief period of time to allow shorter-lived radionuclides to decay; then it is discharged into a system that allows dilution of the remaining radioactivity. Discharge into rivers, lakes, and oceans provides ample dilution of the radioactive waste to below the maximum permissible limits. It is in this manner that waste containing zinc-65 is handled.

When it is discharged from a reactor, zinc-65 is a soluble cation, Zn^{+2}. If discharged into a river, it becomes associated with particulate matter in the water. Studies of the transport of zinc-65 by the Columbia River from Hanford reactors to the Pacific Ocean reveal that changes in physical form and concentration occurring in transit are caused primarily by adsorption on

suspended sediments. Most of these suspended particles settle in the protected and slack water regions of the river. Perkins et al.,[1227] sampling Hanford reactor effluent water from the Hanford reactors and river water at three downstream river locations, demonstrated an increased percentage of zinc-65 in particulate phase from 1.8% at the point of discharge to 14% at Pasco, Washington (56 km downstream) and 64% at Hood River, Oregon (336 km downstream), to 76% at Vancouver, the third point (448 km downstream). They also observed that total zinc-65 concentration at downstream locations decreased, except during spring and early summer when flooding caused scouring of the river bed and resuspension of the deposited radioactivity.

Annual samples of average metal concentrations from 1966 to 1970 taken at Richland, Washington and Bonneville Dam (48 and 384 km below the Hanford reactors, respectively) also showed that zinc-65 in the water decreased with distance because of settling (see Table 4-18).[440a] Because all but one of the production reactors were closed during the time tests were conducted, the zinc concentration diminished from year to year.

As river water mixes with ocean water, the amount of zinc-65 activity associated with particles decreases as salinity of the water increases (see Figure 4-6).[492] This effect and the distribution of dissociated zinc in ocean water have been investigated by the Laboratory for the Study of Radioactive Contamination of the Sea at Fiascherino, Italy. The laboratory has reported that 10-20% of the naturally occurring zinc in sea water is in an ionic state, 30-50% is in particulate form, and 40-50% is in a complexed form. If the ionic form of zinc-65 ($^{65}Zn^{+2}$) is added, its distribution among these forms is not equal. Exchange with the ionic and particulate forms is rapid, but even after 2 years, exchange is incomplete with the complexed forms; complexed zinc-65 is not in chemical equilibrium with the ionic and particulate zinc-65.[943]

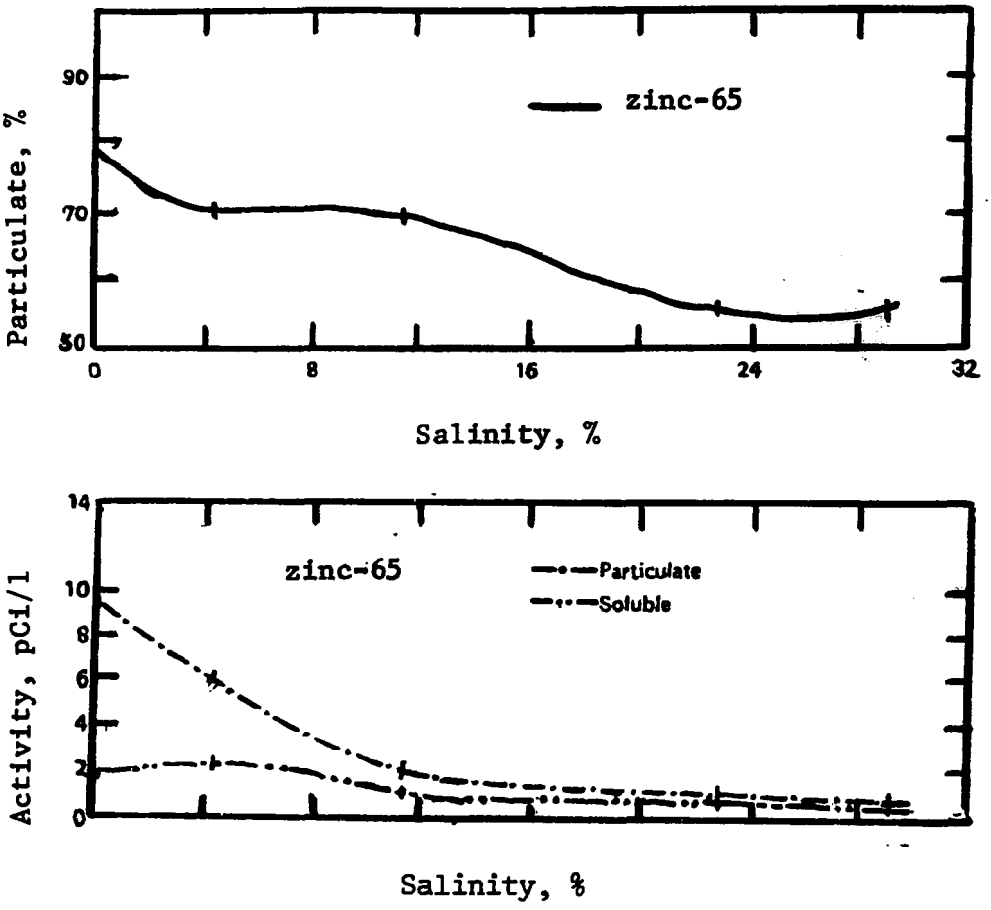


FIGURE 4-6 Soluble, particulate, and percentage of particulate levels in the Columbia River estuary in April 1968. Reproduced from Forster.[492]

Observations of the Columbia River where it merges with the Pacific Ocean have provided additional information on the fate of zinc-65 in estuarine and marine environments. Jennings and Osterberg[792] reported *in situ* radioactivity measurements of estuarine sediments and overlying water; more zinc-65 activity was found in the sediments, and the highest radioactivity was observed in sediments in protected areas of upstream portions of the estuary where fine-grained silts and clays would tend to settle from suspension. Gross[570] and Osterberg,[1181] comparing estuarine sediments with offshore sediments, found that radioactivity decreased rapidly with distance from the mouth of the river and the surface of the water.

This distribution of zinc-65 was further evidenced by radioactivity levels in birds, fish, crustaceans, mollusks, algae, and plankton from along the Washington and Oregon coasts.[1738] Concentrations were highest in samples at the mouth of the river, and they decreased very rapidly with distance from the river mouth, as seen in Figure 4-7. Kujala[883] analyzed 132 samples from 5 species of Pacific salmon and noted distinct differences in radioisotope concentration by species and area of capture. Chinook, chum, and pink salmon that spend most of their ocean life in the Bering Sea and the central north Pacific had zinc-65 activity less than 2.0 pCi/g, while Chinook and coho salmon that live within the Columbia River plume areas had levels as high as 49.2 pCi/g and 59.3 pCi/g, respectively. The highest level, 81.9 pCi/g, was found in Chinook salmon near Eureka, California, which has a nuclear power plant that disposes low-level wastes into the Pacific. Samples of sockeye salmon did not show any increase in zinc-65 activity close to the Columbia River; since these fish are thought to spend most of their ocean life beyond the influence of the Columbia River, they would not be expected to be appreciably influenced by zinc-65 from the river.

Radionuclides in the Columbia River water have been used as convenient tracers to study the dispersion of the river water itself in the Pacific Ocean.

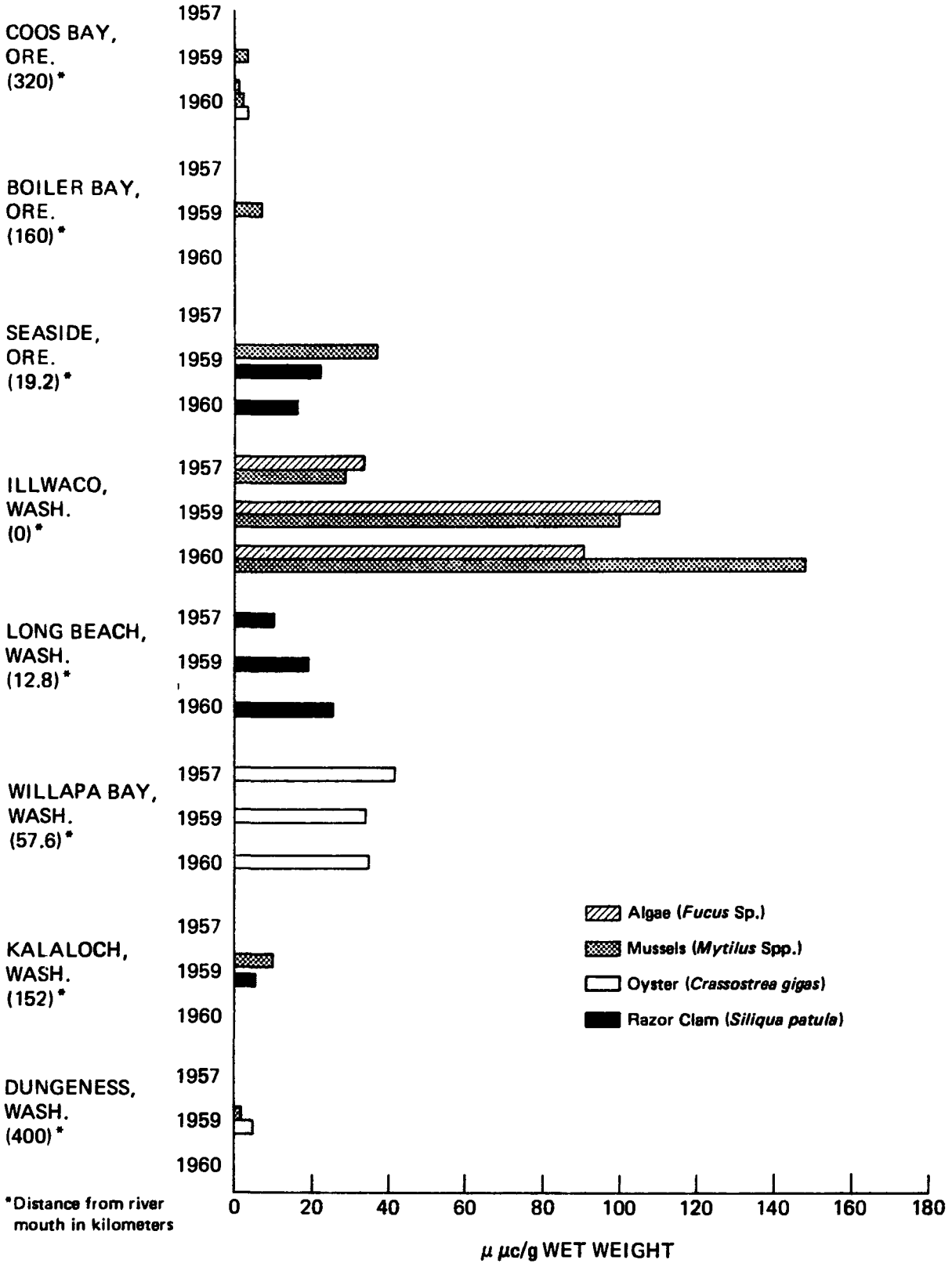


FIGURE 4-7 Concentration of zinc-65 in marine organisms near the mouth of the Columbia River. Reproduced from Watson *et al.* 1738.

Since zinc-65 is taken up by most aquatic organisms and is usually the predominant γ-emitting radioelement found in biota samples, mapping the distribution of zinc-65 by analysis of samples of marine organisms has been particularly useful as an indication of the dispersion pattern of the river water. Such studies[492,1738] have shown that the dispersion follows two seasonal patterns: in summer, because of winds from the north, the flow is offshore toward the southwest as far as 320 km; and in winter, the flow is inshore, north of the river mouth along the coast of Washington, because of the southerly winds.

Research on the distribution and levels of zinc-65 has been prompted by a concern for zinc-65 because it is radioactive, not because it is zinc. Although levels of zinc-65 discharged in effluent are below permissible levels, the potential exists that the radioactivity may be brought back to man in dangerously high levels if it becomes concentrated through food chains. Many studies have been undertaken to evaluate this possibility, including studies using controlled food chains,[72] experiments with oysters raised in reactor effluent,[1397] and surveillance of agricultural products and pastures irrigated with Columbia River water, of the livestock grazing on the pasture grass, and of the humans consuming the produce and livestock.[1228,1229]

The results of these projects and of the Environmental Surveillance Program,[499] aimed at evaluating the radiation dose to man from radionuclides released to the environment, have concluded that although zinc-65 from reactors is detectable in the river and ocean waters and in many species of fish and shellfish, the quantities that reach people are very small and the radiation exposure that results is well within acceptable limits (see Table 4-19).[1643a]

TABLE 4-19

Permissible Levels[a] for Nonoccupational Exposure (Unrestricted Areas)[b]

Type of Exposure	Air, μCi/ml	Water, μCi/ml
Soluble zinc-65	4×10^{-9}	1×10^{-4}
Insoluble zinc-65	2×10^{-9}	2×10^{-4}

[a] Atomic Energy Commission Standards.

[b] Data from U.S. Atomic Energy Commission.[1643a]

CHAPTER 5

ZINC IN PLANTS

AQUATIC PLANTS

Aquatic plants, especially algae, have been used extensively to investigate the role of zinc in plant metabolism. However, much of the research involved laboratory investigations and less is known about zinc in aquatic plants in natural systems. The research on aspects of zinc in aquatic plants other than metabolism has been largely devoted to the fate of zinc-65 as a radioactive contaminant in water systems. Hence, much of the available literature deals with zinc-65 in aquatic plants. A requirement for zinc in the alga Stichococcus bacillaris was demonstrated in 1926,[421] and the metal is now considered to be required by all algae for normal growth and development. Besides the continuing research on metabolic functions and uptake of zinc-65, work is now being done on the ability of aquatic plants to concentrate zinc by several orders of magnitude from ambient waters. Interest has developed in using aquatic plants to remove zinc from contaminated waters as well as in the toxicity of zinc to the plants. Vinogradov[1695] demonstrated that plants living in the ocean remove zinc from the water to produce zinc levels in the ocean which are more acceptable for the normal growth of other organisms.

Zinc Uptake and Concentrations

Uptake. There are two separate mechanisms of zinc uptake by aquatic plants. These are sorption processes (adsorption, absorption, and ion exchange) and metabolic assimilation. Both mechanisms may exist in the same organism, but the pathway utilized often is determined by metabolic requirements or environmental conditions.

Evidence was provided that the uptake of zinc-65 in algae was directly related to the rate of photosynthesis and metabolically controlled.[63] The involvement of adsorption processes in zinc uptake by algae was confirmed when the green alga Golenkinia paricispina was used to show that zinc uptake involved ion exchange sites created by the photosynthetic removal of carbon dioxide.[62] Living and freshly killed algae were placed in the same concentration of zinc-65 and zinc uptake was determined by zinc accumulation in cells over time. Dead cells accumulated more zinc-65 than the living algae. These results were confirmed in studies of freshwater phytoplankton and periphyton.[345,346] Such results indicate that sorption mechanisms accumulate more zinc than is necessary for normal metabolic functions. Gutknecht[590] suggested that metabolic zinc uptake by algae may be a secondary process that occurs only after sorption has taken place. Much of the zinc sorption takes place in the cell walls of aquatic plants.[1452] The sorption processes are probably responsible for the large concentrations of zinc which are found in algae and other aquatic plants when compared to the zinc concentration of the ambient water.

Uptake of zinc by aquatic plants is influenced by the structures in the plants. Algae are often covered by organic excretions that can function as ion exchangers.[146] Because of their suspended state, planktonic algae and free-floating vascular plants can take up zinc from the water but not from sediments. Benthic algae or rooted vascular hydrophytes which are submerged in water may take up zinc from either the sediments or the ambient water. Sediments are usually several orders of magnitude higher in zinc than the ambient water, but the relative importance of sediment and solution in supplying zinc to rooted aquatic plants is unknown.

Concentration. The presence of zinc in aquatic plants has been established for some time.[123,646,1695] Some of the earliest analyses of aquatic plants which showed traces of zinc were conducted in 1919.[318] It was variously demonstrated that the concentration of zinc in brown seaweed (Fucus vesiculosus) varies with the concentration in the seawater.[123,196,591,1815] This observation was confirmed for another brown seaweed, Laminaria digitata.[187,188] Similar results had been obtained earlier.[275]

Bryan,[187] in studies with Laminaria digitata, found 2.2 mg zinc/l of seawater and a growing tissue concentration of 5.4 mg zinc/g, resulting a concentration ratio $\left(\frac{\text{mg zinc/g tissue}}{\text{mg zinc/g ambient solution}}\right)$ of zinc between plant and seawater to be 2,455. Earlier studies by Black and Mitchell[123] produced lower concentration factors, listed in Table 5-1. In a review of mineral nutrition of algae, O'Kelley[1163] reported zinc-65 concentration factors in marine algae ranging up to 1,200. However, concentration ratios for zinc-65 in Columbia River plankton were reported to range from 300-19,000.[344a] Similar high concentration factors were reported for freshwater algae.[649] In later research with brown seaweed, a tissue concentration factor of 3,240 for zinc was reported.[196] Bowen[146] cited ratios found by Russian workers who studied the uptake of zinc-65 by 32 species of algae, bryophytes, and vascular hydrophytes. The concentration factors usually ranged between 2,000-4,000 in tissue as compared to ambient water, where concentrations were low. Chapman et al.[259] found similar conditions determining zinc concentration factors. Boyd[149] found that certain aquatic plants concentrated zinc from the environment in early stages of growth, but uptake decreased as the growing season progressed. Boyd's work does not necessarily disagree

TABLE 5-1

Concentration Factor, or Ratio of Zinc Content in Algae to Zinc Content in Seawater[a]

Species	Concentration Factor $\frac{\text{mg zinc/g tissue}}{\text{mg zinc/g solution}}$
Pelvetia canaliculata	1,000
Ascophyllum nodosum	1,400
Fucus vesiculosus	1,100
Fucus serratus	600
Laminaria digitata frond, Atlantic Bridge	400
Laminaria digitata stipe, Atlantic Bridge	600
Laminaria digitata frond, Ardencaple Bay	1,000
Laminaria digitata stipe, Ardencaple Bay	900

[a]Data from Black and Mitchell.[123]

with the research denoting a continued uptake throughout the growing season, because variations in uptake rate within a specific growing season were seldom determined. That aquatic plants can concentrate zinc from ambient water is well documented.

The content and concentration factor for zinc in algae grown under similar conditions was shown to be species-dependent by Gutknecht.[591] How concentrations of zinc may vary by species is illustrated in Table 5-2. Much of the difference in zinc concentration factors by aquatic plants is caused by differences in the ion exchange properties of their surfaces.[146] One of the primary sites of zinc binding in aquatic plants is in the cell wall.[1452] In studies with Chlorella fusca, Matsku and Broda[991] concluded that some zinc-65 taken up by the organism can be removed by exchange with unlabeled zinc, although another fraction is more tightly bound. Exchangeable zinc is probably in the free space, whereas the bound zinc is in the protoplast. There are probably marked differences in the metabolic assimilation of aquatic plants as well as in their ion exchange properties. Zinc concentrations assimilated by freshwater and marine organisms reported in the literature before 1964 have been summarized,[1257] and other researchers have studied zinc concentration in aquatic plants and reported results similar to those of Matsku and Broda.[105,512,908,1339,1815]

The seasonal changes in zinc concentrations of aquatic plants must be considered when data are compared and interpreted. Table 5-3 contains data on zinc variations by season in several species of seaweed.

Considerable variation in zinc content was evident in some species with different seasonal samplings, but other species contained relatively constant amounts of zinc through all seasons. Sampling techniques probably

TABLE 5-2

Uptake and Concentration of Zinc-65 and Total Zinc in Seaweeds[a]

Species	Zinc-65 Concentration Factor	Total Zinc, mg/kg	
		Fresh Weight	Dry Weight
Ulva lactuca	290	23.80	158.0
Codium decorticatum	30	0.96	17.0
Fucus vesiculosus	3,300	124.00	472.0
Dictyota dichotoma	280	5.70	35.0
Gracilaria foliifera	210	5.83	37.7
Agardriella tenera	395	9.78	91.4
Hypnea musciformis	150	3.54	23.2

[a] Data from Gutknecht.[591]

TABLE 5-3

Seasonal Variation in Zinc Content of Oven-Dried Seaweeds[a]

Sample	Dates Sampled 1/12/49	5/26/49	6/27/50
	mg zinc/kg dry weight		
Pelvetia canaliculata	40	47	90
Fucus spiralis	--	62	no data
Ascophyllum nodosum	103	60	116
Fucus serratus	79	70	63
Fucus vesiculosus	--	60	105
Laminaria digitata frond	99	64	59
Laminaria digitata stipe	no data	62	92
Laminaria coloustoni frond	117	76	--

[a] Data from Black and Mitchell.[123]

account for some of the seasonal variability. However, data have been presented which confirm a seasonal fluctuation in zinc content of Fucus vesiculosus.[196] Knauer and Martin[859a] found that the seasonal variations of zinc in both water and phytoplankton in Monterey Bay, California, were related to the upswelling of the ocean and possibily to the availability of zinc from organic matter on the ocean floor. Other biologic, chemical, and environmental factors that can influence the seasonal variation of zinc in aquatic plants are growth rate, supply of other nutrients, and temperature.

Zinc toxicoses in aquatic plants are distinct possibilities in areas where zinc-contaminated surface waters exist. Complete destruction of aquatic plant communities because of zinc toxicosis in areas associated with mining or processing of ore was reported by Besch and Roberts-Pichette[110] in their study on the effects of zinc mining pollution in the Miramichi River system of northern New Brunswick, Canada.

A laboratory bioassay approach was developed[527,1769] to determine critical levels of zinc that would be potentially toxic to many aquatic plants. The sampling, analysis and correlation studies that have been completed show promise, but variables such as pH, light, temperature, and the presence of other species can influence the reliability of a critical metal level. Dietz[384] conducted bioassays on mosses to assess the trend of water pollution by metals and found that the mosses were considerably enriched by zinc because the water had been contaminated by it. The amount of enrichment also was dependent upon plant species and specific plant selectivity. Gerloff[527] used plant analysis to

evaluate nutrient supplies and growth-limiting nutrients for the aquatic weeds Elodea occidentalis and Ceratophyllum demursum in lakes and streams. The critical concentration of zinc was 8 ppm in the second 2.5-cm segment of main branches and laterals of Elodea occidentalis.

Sprague[1540] used a bioassay that established a critical level of 0.75 mg zinc/l of ambient water as a value which caused lethal effects in aquatic plants. The 0.75 mg zinc/l value applied only when zinc was in an ionic form; the presence of nitrilotriacetic acid (NTA) also reduced the toxicity of the zinc to the plants. Therefore, the form in which zinc exists in solution is an important factor in evaluating potential zinc toxicity to plants.

Tierney[1615] did not find a clear relationship between concentration of zinc in Elodea anadensis and Ceratophyllum demursum and the zinc content of the ambient waters, and he observed that more detailed field and laboratory studies were needed to identify the specific factors which control availability, uptake, and accumulation of zinc by aquatic plants.

Factors Affecting Zinc Uptake and Accumulation

Temperature. Variations in temperature significantly influence the metabolic rate of aquatic organisms.[366] Organisms which obtain zinc through food consumption were shown to increase their zinc-65 accumulation as their rate of food consumption increased at elevated temperatures.[338,637,1332] Similar increases in zinc uptake by aquatic plants can be expected as rates of metabolism increase with increasing temperature of ambient water.

pH. Zinc uptake and accumulation by algae is dependent upon pH.[590,592] Gutknecht found accumulation of zinc-65 by Ulva lactuca to be twice as high at pH 8 as at pH 7 and three times as high at pH 9 as at pH 8. These data and later research, in which zinc uptake was found to follow the Freundlich adsorption equation,[591] demonstrated that pH-dependent adsorption exchange was an important mechanism of zinc uptake by aquatic plants.[183] Brungs observed that the solubility of zinc-65 decreased with increasing pH in freshwaters. These observations were confirmed by the calculation of solubility products, but the precipitation of zinc would be low enough to provide adequate zinc for aquatic plant growth and development at near neutral pH values.

Ambient concentration of zinc. Accumulation of zinc in aquatic plants was found to be directly related to the amount in the ambient water.[123,187, 188,196,591,1815] The log of the zinc concentration in Chlorella cells was discovered to be proportional to the log of the zinc concentration in the medium over a hundredfold range in concentration of zinc.[860] Halving the ambient concentration of zinc-65 available to Chlorella resulted in a 50% decrease in uptake rate and total amount accumulated.[345] Previously Bachmann[62] and Gutknecht[592] had reported similar results with certain other algae species, and similar relationships have been demonstrated for mercury in vascular aquatic plants.[392]

Competing ion. The presence of other cations in waters containing zinc have been shown to reduce the amount of zinc taken up by aquatic plants.

Bachmann[62] found that zinc-65 uptake in the green algae, Golenkinia, was reduced when concentrations of calcium, magnesium, potassium, and sodium were increased. Cushing and Rose[345] reported that zinc-65 uptake by periphyton was reduced 50% when the amount of magnesium in solution was doubled. Also, doubling the stable zinc concentration decreased the zinc-65 uptake by 25% (probably caused by isotopic dilution). The competition between zinc-65 and other cations for available binding sites on the plants is responsible for such occurrences.[345]

Light. If minimum requirements for photosynthesis are met, increased light intensity will cause little increase in zinc uptake or accumulation.[590,592]

Functions of Zinc

Zinc functions primarily in plants as a metalloenzyme.[1296] Several zinc-requiring dehydrogenases, proteinases, and peptidases have been identified in various organisms, including aquatic plants. Price[1295] reported that several dehydrogenases were sensitive to zinc deficiency in Euglena gracilis. Therefore, the metabolism of aquatic plants can be influenced markedly by changes in the availability of zinc. Data indicate that zinc deficiency rapidly reduces the amount of RNA as well as the ribosome contents of cells.[1296] Another function for zinc in plant metabolism may be as a stabilizer of the cytoplasmic ribosomes of Euglena gracilis.[1289,1296] It also has been suggested that zinc aids in auxin B production, but a definite connection between zinc and indolacetate has not been determined.[1296] For additional information on the metabolic role of zinc in connection with metalloenzyme activity, see Chapter 9.

TERRESTRIAL PLANTS*

Reports of the use of zinc as a nutrient in plant fertilizer appeared in the literature as early as 1912,[790] and Brechley[157] described zinc deficiency in higher plants in 1914. That zinc was essential to plant life was established by Sommer and Lipman[1523] when they reported that zinc was required for three plant species. Zinc was discovered to be necessary for trees when zinc deficiency was identified on fruit trees in California,[255] and when pecan rosette (a malformation of the new leaves of growing tips) in the South was found to be caused by zinc deficiency.[14] Knowledge of the nature of zinc made the identification of many nutritional problems in higher plants possible. Zinc deficiency is now the most common micronutrient deficiency in the United States.[938] Using zinc fertilizer to improve crop growth also is well established. Although it is possible for zinc toxicity to be a problem to plants, zinc deficiency in plants is far more likely than toxicosis. Crops that are sensitive to zinc deficiency are citrus and deciduous tree fruits, pecans, pine (in Australia), grapes, kidney beans, hops, soybeans, corn, lima beans, flax, castor beans, and onions. Cotton, potatoes, tomatoes, alfalfa, clovers, sorghum, sudan grass, and sugar beets are mildly sensitive to zinc deficiency, and peas, small grains, peppermint, asparagus, mustard and other crucifers, forage grasses, safflower, and carrots are not sensitive to it.[1686]

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*The research discussed in this section appeared in the literature before April 1975.

Availability of Zinc in Soils

Climatic and soil conditions responsible for zinc deficiency in crops in the United States were reviewed in detail by Lucas and Knezek.[956] Zinc deficiency in European countries was reviewed earlier.[1387] Some important factors associated with or contributing to zinc availability in soils are summarized below. Further details may be found in two reviews.[938,956]

Soil low in zinc reserves. Quartz is low in zinc, so sandy soils often contain only 10-30 ppm total zinc.[938] A similar observation has been reported for peat and muck soils, where zinc in the plant root is separated from the mineral reserves below the organic surface layer. Under acid conditions, zinc may be rapidly leached from the soil where rainfall is great.[956]

High soil pH. Most disorders in plants caused by zinc deficiency occur in calcareous soils with a pH of 7.4 or higher, because the solubility of zinc decreases as soil pH increases. The total zinc content of calcareous soils is often equal to or higher than that of noncalcareous soils. In addition to precipitation at high soil pH values, adsorption of zinc by carbonates may contribute to low availability of zinc to plants on certain calcareous soils.[939,956]

Limited root zones. Zinc deficiencies are frequently found in soils with restricted root zones. They may be caused by hardpans, high water tables, or other factors. Equipment passage may compact soil and cause zinc deficiency in certain agricultural soils.[956] The root development of rice

grown on flooded and nonflooded soils in Alabama determined the amount of zinc available to plants.[535]

Microbial fixation and low soil organic matter. Zinc deficiency in beans and corn on calcareous soils is often observed on old corral sites and barnyards and has been attributed to the rapid growth of microorganisms that may tie up available zinc.[956] Zinc deficiency in corn is sometimes more severe when it has been planted after a crop of sugar beets than when planted in soil used for corn or many other crops.[938] DeRemer and Smith[378] observed that plowing down sugar beet tops reduced available zinc to the succeeding bean crop. Therefore, organic matter or soil microorganisms help determine the zinc fixation by soils. Zinc deficiencies are often reported in areas where surface soil containing organic matter has been removed.[572] Lindsay[938] concluded that deficiencies increased because the exposed subsoil was lower in organic matter and higher in pH and carbonate than the surface soil. Indeed, a close correlation between soil organic matter and extractable zinc has been well documented.[487, 794,1623] More observations of zinc deficiencies were reported in Michigan where the surface soil was disturbed and the subsoil exposed.[956]

Soil temperature and moisture. Cool, wet spring weather often aggravates or induces zinc deficiency in field crops.[956] Limited plant root growth, which brings about a limited root feeding zone and reduced microbial activity so that zinc is not released from organic matter during the cool, damp weather, may cause such deficiencies. Wallace, et al.[1719] have found

that zinc uptake increases with temperature. Bauer and Lindsay[86] concluded that decreased solubility of zinc in the soil, rather than a biologic effect, was the primary reason for more pronounced zinc deficiencies in cool weather. Additional research[523] supported those results. Soil moisture and aeration influence zinc availability through indirect soil reactions[432] and root metabolism.

High phosphate levels in soil. A phosphorus-induced zinc deficiency has been widely reported.[131,435,523,977,1121,1384,1465,1686] Application of phosphate fertilizer and high phosphorus levels in soils have been linked to zinc deficiency problems for many years.[78,1611] Since zinc extractable from soil is not decreased by the addition of phosphorus to soils, the formation of insoluble zinc phosphate compounds in soils is probably not the cause of a phosphorus-induced zinc deficiency.[939,1168] The deficiency may result from the formation of an iron phosphate at the root surface that excludes zinc uptake where soil zinc levels already are low.[432]

Influences of nitrogen in soil. The acidifying influences of nitrogen fertilizers -- which make zinc solubility greater and possibly increase the cation exchange capacity of roots when the nitrogen supply increases -- have been cited as causes of enlarged zinc uptake in plants.[132,396,939,1687]

Movement of Zinc to Plant Roots

Zinc is believed to move through the soil to plant roots by mass flow (convection) and diffusion.[938] Mass flow is the movement of nutrients

along with the soil solution, whereas diffusion is the movement of nutrients through the soil solution because of a concentration gradient.[1775] (Olsen and Kemper[1169] provide a detailed review on movement of nutrients to plant roots.) Various workers[73,74,238,290,371,425,426,434] have shown that diffusion gradients are formed between the soil solution and plant roots and that most zinc movement to plant roots is by diffusion. Warncke and Barber[1733-1735] have explained how the rate of zinc diffusion in soil is controlled. Soil moisture, bulk density, and zinc adsorption are all important in determining final zinc diffusion rates. Wilkinson *et al.*[1776] have reported that tripling the transpiration rate in wheat seedlings did not yield an increased uptake of zinc, and concluded that zinc movement to plant roots was independent of mass flow and dependent upon diffusion. These results were confirmed for several species.[607] The movement of zinc to plant roots is dependent upon soil solution concentration and the ability of soil to replenish zinc removed from solution.[938] Lindsay and Norvell[941] reported that increasing soil pH decreased solubility of zinc in soils, and Lindsay[939] stated that the diminished zinc concentration and subsequent reduced concentration gradient would reduce zinc availability and uptake by plants. Melton *et al.*[1028] have confirmed the influence of soil pH on zinc diffusion and found a limited influence of phosphorus on zinc diffusion.

The effects of chelates upon zinc solubility and availability to plants have been well documented.[170,174,175,212,372,425,426,574,712,938,941,1149,1717] Chelating agents increase the solubility of zinc in soils and increase the movement of zinc to plant roots by both mass flow and diffusion.[938]

The ability of the plant root to compete with the chelating agent for the zinc will determine if the actual amount of zinc uptake by plants will be increased or decreased from chelation. Adding chelating agents to soils will usually increase zinc availability to plants because the solubility is increased; but the addition of a chelating agent to nutrient solutions may reduce the uptake of zinc that is already in solution.[938]

Zinc Uptake, Translocation, and Concentration by Plants

Uptake. Zinc absorption by plants has been reviewed,[938,1078] and factors influencing plant absorption of zinc have been identified.[696,1554] Ranges of zinc uptake from 2-4,000 ng/day on a fresh weight basis have been reported.[239,1428] Hewitt[695] reported optimal levels of zinc for plants growing in nutrient solution to range from 0.3-3.0 μM, Carroll and Loneragan[238] observed that a variety of plants grew well at 0.01 μM with maximum growth at 0.25 μM zinc. Zinc absorption by sugar cane has been investigated with similar findings.[147]

Considerable uncertainty still exists about the roles of active and passive mechanisms in zinc uptake by plants.[938,1078] Conclusions of workers who failed to separate passive exchange absorption from active cellular accumulation are the source of much conflict.[166,441,1078,1317, 1318,1428] For example, some researchers of zinc translocation found that xylem exudates from tomatoes and soybeans contained higher concentrations of zinc than did the nutrient solution in which the plants were grown.[27,1617] Such results suggest an active accumulation component in the plant if it is assumed that the zinc in the xylem exudate is present in a form similar

to that in the external nutrient solution. Most of the evidence based on study of metabolic inhibitors points toward an active accumulation component crucial to zinc uptake.[536]

Translocation. Zinc is translocated from the roots to the tops of plants through the xylem, and a limited amount of zinc is retranslocated from the leaves through the phloem.[938,1617,1618,1721] (For a review of the translocation in plants, see Tiffin.[1618]) It is intermediately mobile within plants compared with other micronutrients.[938] Zinc is localized in plant roots when zinc supply is normal,[238] but it moves from roots to plant tops when in limited supply.[984,1340] Also, some plants increase the efficiency of the zinc they have stored by redistributing it from seeds to leaves and from older to newer plant tissue.[200,938,1088,1618]

Olsen[1168] reviewed the interactions of zinc and other elements in plants. Generally, zinc and phosphorus interactions reduce the uptake and translocation of zinc by plants.[111,113,169,827,1084,1502,1559,1604] Also, a high level of available nitrogen often causes increased zinc deficiency. Much of this deficiency was attributed to increased formation of zinc-protein complexes in plant roots that inhibited normal translocation to plant tops.[1168] Excess copper or iron in a plant also will bring about intensified zinc deficiency. The influence of zinc in suppressing copper uptake and translocation by plants usually is greater than the suppressive effect of excessive copper upon zinc translocation.[263] Iron levels were reported to be high in zinc-deficient sweet corn.[786] The application of phosphorus decreased zinc concentrations in plants and

plants low in zinc were high in iron. Ambler and Brown[26] have observed that lowering zinc concentration in solution has increased the translocation of iron and phosphorus into bean leaves. Because the iron and phosphorus consistently increased in bean plants, the phosphorus evidently did not inhibit iron translocation. Similar findings have been reported for corn,[1737] and decreasing the amount of zinc applied increased the phosphorus uptake nearly sevenfold in leaf tissue, but only threefold in stems and roots.

Zinc uptake and translocation in tomato, soybean and squash plants were increased when the salt concentration of the growth medium was increased.[960] Hence, there may not be an antagonistic relationship between soluble salts and zinc in nutrient culture. Nor has additional research on iron, manganese, and zinc interactions (using isotopic techniques) been conclusive.[452] Macronutrient cations, as well as copper and hydrogen, suppressed zinc uptake and translocation by wheat seedlings, but iron and manganese had no effect on zinc uptake.[261,262] Therefore, the evidence for zinc interactions with various elements is not solid and varies according to plant species and other minerals present.

For example, Brown *et al.*[171] reviewed the differential susceptibility of various plant genotypes to zinc deficiency. A study of zinc in temperate crops and pasture grasses revealed that species differ in their ability to take up and utilize zinc.[540] Similar findings have been reported for navy beans,[431,803,1258,1317] oats,[209] rice,[534] soybeans,[1716] sugar cane,[128] and wheat.[1492] A definite genetic basis for zinc uptake, distribution, and utilization by corn plants was confirmed by others.[603,984]

Concentration. Sauchelli[1417] and Jones[800] summarized ranges of zinc concentration in plants. When zinc drops to levels below 20 ppm dry weight in leaves, deficiencies may occur. The normal concentration range is 25-150 ppm. Zinc and toxicoses often occur when levels exceed 400 ppm.[800] Zinc concentration ranges for several specific plant species have been reported.[85,258,540] The concentration of zinc in plant tissue usually is greatest in young plants; it decreases because of dilution and redistribution as the tissues grow and mature.[550,799,1088,1341] Crops were classified into three groups by Viets et al.[1689] according to their zinc concentration. Table 5-4 sets forth the variety of zinc concentrations possible which do not produce visible symptoms of deficiency or toxicosis.

Zinc Disorders in Plants

Deficiency. Zinc deficiency symptoms have been described in detail for several plants,[1417] and zinc deficiency and its control have been reviewed.[1612] Because zinc is intermediate in mobility within the plant, a variety of symptoms, including chlorosis, stunting, and necrotic tissue may indicate zinc deficiency. In corn and sorghum plants, chlorotic streaks appear in older leaves. The older leaves of navy beans and soybeans turn yellow, and the entire plant becomes stunted and fails to produce a normal crop when the deficiency is severe. In citrus, peach, pecan, and tung trees, irregular chlorosis and bronzing occurs and the new growth becomes stunted. Severe deficiency may kill the leaves and twigs and the growing tip may be deformed, exhibiting a symptom known as "rosetting."

TABLE 5-4

Zinc Content of Crop Plants[a]

Crop Plant	Zinc, ppm				
	Deficient	Low	Normal	High	Toxic
Corn leaves, vegetative stage	0-10	11-20	21-70	71-150	150
Soybean leaves, vegetative stage	0-10	11-20	21-70	71-150	150
Wheat, barley oats, vegetative stage (7.5-30 cm)	0-10	11-20	21-40	41-150	150
Cotton, vegetative stage	--	--	20-30	--	--
Tobacco, vegetative stage	--	0-20	21+	--	--
Sugar beets, vegetative stage	0-10	11-20	21-70	70+	--
Potatoes	--	0-16	17-40	30+	--
Alfalfa tops	0-8	--	--	9-14	--
Grass, vegetative stage	--	--	15-80	--	--
Beans (leaves)	0-20	--	--	--	--
Totatoes (leaves)	0-10	11-20	21-120	121+	--
Citrus (leaves) Florida	0-15	16-25	26-80	81-200	200
Tung (leaves)	0-10	11-26	--	--	--
Apples (leaves)	0-15	16-20	21-50	51+	--
Peaches (leaves)	0-16	17-20	21-50	51+	--
Pears (leaves)	0-10	11-16	17-40	41+	--
Grapes (petioles)	--	0-30	31-50	51+	--

[a]Derived from Sauchelli[1417] and Bishop and MacEachern.[120]

Symptoms of zinc metabolic disorders also are expressed cytologically and morphologically. There are more free amino nitrogen and amides in zinc-deficient plants than in healthy ones.[1267] Inorganic phosphate also is higher in the region outside the stele and in the phloem of deficient stem tissue.[1321] In many plant species, zinc deficiency is shown by interveinal chlorosis[258] because the chlorophyll formation has been disrupted.[1453] According to Seatz and Jurinak,[1464]

> the palisade cells of leaves are larger and transversely divided, rather than columnar; reduction in number of chloroplasts; the absence of starch grains; the presence of oil droplets in the chloroplasts and the presence of calcium oxalate crystals and accumulation of phenolic materials in the leaves.

are morphologic changes that plants undergo when deficient in zinc.

Diagnosis and correction of deficiency. Plant and soil analyses can be useful tools in diagnosing zinc deficiency in many plants. A current review which includes soil testing and plant analysis techniques for zinc has been edited by Walsh and Beaton.[1724] Zinc deficiency usually is suspected if plant tissue concentrations are below 20 ppm zinc, but the critical level varies with species, stage of growth, and plant part sampled.[1026] Levels of zinc in solutions obtained from soil extractions with acids or chelating agents have been correlated with the occurrence of zinc deficiency. Usually, about 50 ml of solution is used to extract zinc from 10 g of soil. Use of 0.1 N hydrochloric acid has been accepted for several years, but solutions containing ethylenediaminetetraacetic acid (EDTA) and diethylenetriaminepentaacetic acid (DTPA) have also been used. Critical zinc levels in the extracting solution that have been related to zinc deficiency in plants range from 0.5–2 ppm zinc.[21,1724] Hag and Miller[631] have compared four

zinc extractants on 65 soils and found EDTA and DTPA extractions to be most useful in predicting zinc deficiency. But Evans et al.[445] found few differences in correlations between zinc uptake and soil zinc extracted with several extractants. The reliability of soil tests to predict zinc availability in soils is dependent upon many soil and plant factors and no single extractant works best in all situations.[1623] Table 5-5 summarizes recommendations of zinc fertilizer applications on specific crops by state.

With most plants, zinc deficiency can be overcome by soil or foliar application of zinc salts.[927,955] Zinc sulfate or finely powdered zinc oxide are the inorganic zinc salts usually applied. However, Wallace and Romney[1718] have used zinc sulfate, zinc NTA and zinc EDTA with great success; seven different zinc sources were evaluated extensively and found to be effective during the first few months after application. However, the zinc apparently reverts to forms of equal availability after 1-3 yr. Banding the zinc fertilizer 2.5 cm to the side of and 5.0 cm below the seed provides the most efficient use per pound of zinc by responsive crops,[801,802] and banded and broadcast applications of zinc to soils for crop response have been successfully used.[963,1693] Zinc has been applied to corn seed at planting time without injury,[980] and the plants have responded to the zinc fertilizer, but many crops may suffer from salt toxicosis during germination. Applying zinc in the form of synthetic chelates increased zinc efficiency from two- to fivefold compared to zinc sulfate.[128,1693] But certain zinc chelates are relatively soluble and can be leached out of the plant root zone when the soil is excessively moist.[892] Most crops use less than one kilogram of zinc per heetare of soil per year.

TABLE 5-5

Recommendations for Applying Zinc Fertilizer by State[a]

State	Crops	Recommendations for Zinc
Alabama	pecan	1.8 - 2.25 kg $ZnSO_4$/tree/yr
	corn	11.25 kg $ZnSO_4$/ha
California	all tree crops and field crops	tree: 1.02 - 2.96 kg $ZnSO_4$/100 liter water field: 11.25 - 22.5 kg $ZnSO_4$/ha
Colorado[b]	corn	5.63 - 11.25 kg $ZnSO_4$/ha
Florida	tree crops	360 g $ZnSO_4$, plus 120 g lime/100 liter water
	vegetable and field crops	90 - 225 g ZnO every 4-5 yr
	ornamentals	90 - 270 g ZnO every 4-5 yr
Georgia	pecans	0.9 kg $ZnSO_4$/cm trunk diameter
	peaches	0.9 - 1.35 kg $ZnSO_4$/tree
	corn	11.25 - 22.5 kg $ZnSO_4$/ha
Indiana	corn	0.9 kg $ZnSO_4$/ha applied as plow down
Iowa	corn, soybeans	11.25 - 22.5 kg $ZnSO_4$/ha
Kansas	corn, sorghum	11.25 - 16.88 kg $ZnSO_4$/ha
Kentucky	corn	4.5 kg $ZnSO_4$ in row
Louisiana	citrus, pecans, tung	360 g $ZnSO_4$, plus 120 g lime/100 liter water
Maryland	corn	11.25 kg $ZnSO_4$/ha
Michigan	beans	2.25 - 4.5 kg $ZnSO_4$/ha

TABLE 5-5 (Continued)

State	Crops	Recommendations for Zinc
Minnesota[b]	corn	11.25 kg $ZnSO_4$/ha
Mississippi	pecans, tung, corn	2.25 - 4.5 kg $ZnSO_4$/tree 11.25 kg $ZnSO_4$/ha
Montana[b]	cherries, apples	16.88 kg $ZnSO_4$/ha
Nebraska[b]	castor beans, corn, fruit, sugar beets, soybeans	5.63 -22.50 kg $ZnSO_4$/ha
North Carolina	peaches	44.2 g $ZnSO_4$/100 liter water every 3 wk
North Dakota[b]	corn, potatoes	16.88 kg $ZnSO_4$/ha
Oklahoma[b]	pecans	4.5 kg $ZnSO_4$/ha
Oregon[b]	corn, lima beans all tree fruit cherries pear barley	5.86 - 12.38 kg $ZnSO_4$/ha 204 g/100 liter water 1.42 - 1.77 kg $ZnSO_4$/liter water 590 g $ZnSO_4$/100 liter water 16.88 kg $ZnSO_4$/ha
South Carolina	corn pecans vegetables	11.25 kg $ZnSO_4$/ha 4.5 - 5.4 kg $ZnSO_4$/mature tree 5.63 kg $ZnSO_4$/ha
Tennessee	corn	11.25 - 22.5 $ZnSO_4$/ha
Texas	pecans	foliar: 360 g $ZnSO_4$/100 liter water
Utah[b]	fruit	11.25 kg $ZnSO_4$/ha

[a]Derived from Sauchelli.[1417]
[b]Deficient levels are less than 16-20 ppm zinc in plant, oven-dry basis.

Thus, 1.35-4.5 kg/ha inorganic zinc salts/yr gradually builds up the zinc reserves in the soil. Adding 28.13 kg zinc/ha as zinc sulfate just once provides an adequate zinc reserve for 10 years of maximum crop production on severely zinc-deficient soil.[1693] Similar residual effects of zinc fertilizer have been reported.[129,652,938,1432] Foliar application of zinc to plants, often used to provide supplemental zinc, is usually imposed at rates of 1.13 kg zinc/ha/application.[927,1062] It also has been reported[90] that certain surfactants enhance foliar absorption of zinc. Once absorbed, zinc is not easily leached from vegetative parts of plant tissues.[233,1795]

Toxicosis. It is well established that excess zinc can be toxic to plants.[130,133,256,258,519,913,1095,1320,1546] Staker and Cummins[1546] found zinc toxicosis in onions, spinach and potatoes in some peat soils in New York. The soils had accumulated several thousand ppm zinc (normal soils contain 10-300 ppm).[97] However, few areas of natural zinc toxicity exist. Toxic amounts of zinc usually occur in acid soils or in contaminated areas such as mine spoil banks, industrial areas, waste disposal sites, or areas of mine seepage. Zinc was toxic to corn and cowpeas when added to soil at rates of 560-1,120 kg zinc/ha.[519] A concentration of 50 ppm zinc in Sanilac field beans can be poisonous; toxicosis at this concentration was produced mainly with acid soils.[1027] Zinc toxicosis has been reported in several plants in greenhouse studies when excessive levels of zinc were added.[256] Boawn[130] and Boawn and Rasmussen[133] have investigated potential zinc toxicosis in many plants, and their results are provided in Tables 5-6

TABLE 5-6

Zinc Concentration in Leafy Vegetables Grown at Normal and Excessive Rates of Zinc Fertilization[a]

Crop	Variety	Sample Description	Zinc Treatment, kg/ha						
			0	11	55	112	224	448	896
			ppm						
Head Lettuce (Lactuca sativa, var. capitata)	Imperial 847	Market size heads	38	45	64	94	144	165	248
Leaf Lettuce (Lactuca sativa, var. crispa)	Grand Rapids	Market size plants	38	46	64	125	157	239	269
Romaine Lettuce (Lactuca sativa, var. longifolia)	Parris Island	Whole plant before heading	32	40	56	78	108	146	179
Romaine Lettuce (Lactuca sativa)	Parris Island	Market size heads	48	50	62	76	100	117	122
Endive (Cichorium endivia)	Green Curled	Market size plants	32	38	73	142	247	308	343
Parsley (Petroselinum hortense)	Extra Curled Dwarf	Market size plants	58	50	86	107	188	296	438
Swiss Chard (Beta vulgaris, var. cicla)	Lucullus	Market size plants	80	72	153	325	615	704	862[b]
Spinach (Spinacia oleracea)	Improved Thick Leaf	Market size plants	139	119	148	175	240	344	340[b]
Chinese Cabbage (Brassica pekinensis)	Chihli	Whole plant before heading	54	48	68	84	89	112	114
Chinese Cabbage (Brassica pekinensis)	Chihli	Market size heads	46	42	60	71	112	248	389
Mustard (Brassica juncea)	Florida Broad-leaf	Market size plants	32	32	36	43	58	131	364
Collard (Brassica oleracea, var. acephala)	Vates	Rosette of young leaves	33	34	38	42	63	104	366
Cabbage (Brassica oleracea, var. capitata)	Earliana	Market size heads	22	20	23	28	34	54	73
Brussel Sprouts (Brassica oleracea, var. gemmifera)	Long Island Imperial	Market size heads	50	47	56	50	62	73	79

[a]Data from Boawn.[130]

[b]At the indicated rate of zinc fertilization, plants showed normal color but were stunted.

and 5.7. King and Morris[842] reported zinc toxicosis in rye at soil applications of 600 kg zinc/ha in sewage sludge on sandy loam soil, but no toxicosis was observed in Bermuda grass at the same zinc levels.[841] Zinc levels in the plants were in excess of 300 ppm at the highest rate of zinc application. Zinc toxicosis was induced in pine trees on three coastal plain soils of Florida when 200-300 ppm zinc was added to the soil.[1673] Tissue concentrations reached 300 ppm zinc at the highest application rates.

Conversely, Chesnin[267] observed that field experiments in Nebraska involving zinc applications up to 700 ppm zinc for corn on acid and alkaline soils did not bring about toxicosis. However, corn is thought to be moderately tolerant to excessive zinc levels in soil. In Michigan, a single application of 140 kg zinc/ha did not induce zinc toxicosis to navy beans on a calcareous soil.[1693] Nor did additions of 387 kg zinc/ha result in toxicosis in corn, cucumbers, or snap beans grown on a sandy soil in Wisconsin.[1725] Six annual additions of 11.1 kg zinc/ha on a sandy soil in Virginia did not cause toxicosis in soybeans.[979]

Tolerance. Certainly many plant and soil factors influence the susceptibility of various species to zinc toxicosis. However, certain species have a relatively high tolerance to zinc in soils. Antonovics et al.[43] have reviewed the zinc tolerance of various plant species growing upon zinc-contaminated soils. Plant species that can tolerate zinc are: Rumex acetosa, Festuca ovina, Agrostis stolonifera, Agrostis canina, Viola lutea, Alsine verna, Silene vulgaris, Plantago lanceolata, Linum catharticum, Campanula tenuis, Festuca rubra, Holcus lanatus, Anthoxanthum odoratum, Thlaspi alpestre, and Armeria maritima.[43]

TABLE 5-7
Growth Response and Zinc Accumulation by Crops Grown at Excessive Rates of Zinc Fertilization[a]

Zinc added, ppm	Field corn (Zea mays), Idahybrid 330		Sweet Corn, Golden Cross Bantam		Sorghum (Sorghum bicolor), NR-125		Sorghum (Sorghum bicolor), RS-626		Barley (Hordeum vulgare), Trail	
	zinc in tops, ppm	yield decrease %	zinc in tops, ppm	yield decrease %	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%
10	37	0	41	0	34	0	32	0	70	0
100	205	0	255	8	380	10	357	7	220	10
200	314	13	367	12	506	30	571	11	530	16
300	484	20	475	32	748	43	646	50	910	42
400	576	26	695	55	917	62	975	66	1,237	59
500	763	42	713	48	1,029	80	1,140	70	2,112	76
% yield decrease for significant at .05 probability level:		26		29		15		19		15

Zinc added, ppm	Wheat (Triticum vulgare), Gaines		(Phaseolus vulgaris) Field beans, Big Bend		(Phaseolus vulgaris) Snap beans, Yakima		Alfalfa (Medicago sativa), Vernal		Clover (Trifolium pratense), Ladino	
	zinc in tops, ppm	yield decrease %	zinc in tops, ppm	yield decrease %	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%
10	51	0	24	0	21	0	27	0	28	0
100	185	1	66	0	46	11	71	0	81	0
200	345	3	101	0	69	8	97	3	109	2
300	522	18	151	0	111	8	142	0	161	7
400	682	30	213	0	142	14	232	17	202	0
500	909	45	257	10	213	12	345	22	252	9
% yield decrease for significant at .05 probability level:		10		ns		ns		13		ns

Zinc added, ppm	Pea (Pisum sativum), Perfection		Pea, Alaska		Lettuce (Lactuca sativa), New York		Spinach (Spinacia oleracea), Thick leaf		Potato (Solanum tuberosum), Russet Burbank	
	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%
10	37	0	36	0	34	0	72	0	33	0
100	132	0	104	0	96	18	338	0	79	0
200	197	4	166	1	152	4	452	1	125	0
300	285	6	236	9	250	21	640	12	163	0
400	367	7	379	10	390	18	775	19	236	0
500	489	8	522	30	665	31	945	32	327	0
% yield decrease for significant at .05 probability level:		ns		22		27		10		ns

Zinc added, ppm	Potato White Rose		Sugar beet (Beta vulgaris), Monogerm Hybrid		Tomato (Lycopersicon esculentum), Royal Ace	
	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%	zinc in tops, ppm	yield decrease%
10	28	0	39	0	51	0
100	67	2	162	17	150	0
200	95	1	355	14	257	5
300	138	5	509	13	316	8
400	212	0	851	23	381	18
500	346	8	1,067	40	514	26
% yield decrease for significant at .05 probability level:		ns		22		24

[a]Data from Boawn and Rasmussen.[133]

Plants that have been used as indicators of high levels of soil zinc are listed below. Although many plants are sensitive to high soil zinc levels, certain plants can provide vegetative cover on zinc-contaminated soils because of their tolerance for high zinc levels in soils. In Europe, Minuartia verna. Armeria vulgaris (and halleri, elongata, martima, etc.), and Viola calaminaria announce the presence of zinc; in Australia, Tephrosia sp. nov., Tephrosia affin. polyzyga, Polycarpaea synandra var. gracilis, and Gomphrena canescens are such reliable markers that they are used as such in prospecting for heavy metals. Philadelphus sp. is a similar signal in the state of Washington.[43]

The mechanisms of zinc tolerance in colonial bent grass (which is readily tolerant to zinc) and creeping bent grass (which is not) have been well examined.[1237,1633,1634] Similar amounts of zinc entered the tolerant and intolerant plants and similar amounts reached the tops, but the tolerant plants appeared to deactivate zinc. Most of the cytoplasmic zinc in the tops of colonial bent grass existed as a stable anionic complex.[1237] Considerable differences were noted among the physiologic responses of the bent grass species.[1633,1634] The binding of zinc in the cell wall fraction was suggested as the mechanism of zinc tolerance in the colonial bent grass. Species other than bent grass have developed zinc-tolerant ecotypes, but they have not yet been studied in detail.[563]

The relationships between cadmium and zinc in grasses are quite important to human and animal nutrition. Huffman and Hodgson[744] determined the zinc:cadmium ratios in wheat and perennial grass samples from 19 states

east of the Rocky Mountains. No zinc:cadmium ratios appeared to be determined by regions of the country in which the plants grew. These data are summarized in Table 5-8. High levels of zinc may suppress cadmium uptake by plants, but at levels of zinc normally found in soils, suppression does not seem to occur.[256,598,1632] There is evidence that each element may enhance the uptake of the other even when relatively low levels of cadmium and zinc are present in the soil.[256]

Zinc in Plant Metabolism

The specific functions of zinc in plant metabolism are not completely defined, but much progress has been made in the last 30 years. Established functions of zinc in plant metabolism have been reviewed.[1296]

The best defined and most important role of zinc in plants is as an enzyme cofactor.* Carbonic anhydrase was the first zinc metalloenzyme to be identified in plants and considerable research has been and is still conducted on zinc and carbonic anhydrase activity.[55,56,371,415,450,554a,1130,1312,1488,1624,1803] Many metalloenzymes that require zinc have been described.[710] Zinc binds pyridine nucleotides to the protein portion of enzymes and zinc atoms stabilize the structure of yeast alcohol dehydrogenase.[598a] It has been recognized as an essential component of a variety of dehydrogenases, proteinases, and peptidases, as well as the zinc metalloenzyme carbonic anhydrase.[1664] In plants they include alcohol dehydrogenase, glutamic dehydrogenase, L-lactic dehydrogenase, D-glyceraldehyde-3-phosphate-dehydrogenase, glutamic dehydrogenase, D-lactic dehydrogenase, D-lactic cytochrome c reductase and aldolase.

* For more information on zinc and enzymatic activity, see Chapter 9.

TABLE 5-8

Average Cadmium Concentrations and Zinc:Cadmium Ratios Found in Wheat and Perennial Grasses[a]

State	Crop	Number of Samples	Cadmium, ppm	Zinc:Cadmium, wet weight
Alabama	Wheat	-	--	-
	Grass	8	0.17	122
Colorado	Wheat	9	0.19	80
	Grass	2	0.27	80
Connecticut	Wheat	-	--	-
	Grass	7	0.13	187
Georgia	Wheat	1	0.18	94
	Grass	23	0.16	131
Illinois	Wheat	1	0.34	58
	Grass	8	0.21	107
Iowa	Wheat	-	--	-
	Grass	7	0.26	88
Kansas	Wheat	7	0.16	94
	Grass	6	0.15	138
Kentucky	Wheat	1	0.12	108
	Grass	2	0.11	147
Louisiana	Wheat	-	--	-
	Grass	1	0.18	172
Massachusetts	Wheat	-	--	-
	Grass	7	0.13	191
Mississippi	Wheat	-	--	-
	Grass	2	0.44	70
Nebraska	Wheat	5	0.32	55
	Grass	3	0.25	76
New York	Wheat	-	--	-
	Grass	6	0.14	130
North Carolina	Wheat	4	0.22	96
	Grass	4	0.20	133
Ohio	Wheat	-	--	-
	Grass	8	0.17	152
Oklahoma	Wheat	2	0.12	97
	Grass	1	0.14	143
South Carolina	Wheat	-	--	-
	Grass	4	0.18	115
Texas	Wheat	2	0.14	168
	Grass	21	0.15	166
Virginia	Wheat	1	0.25	60
	Grass	-	--	-
Summary of Data from Samples Taken East of the Mississippi				
	Wheat	8	0.22	83
	Grass	79	0.17	139
	Overall	87	0.17	136
Summary of Data from Samples Taken West of the Mississippi				
	Wheat	25	0.20	87
	Grass	41	0.18	137
	Overall	66	0.19	118
Summary of All Data				
	Wheat	33	0.20	88
	Grass	120	0.17	137
	Overall	153	0.18	126

[a] Data from Huffman and Hodgson.[744]

Tomato plants grown in zinc-deficient media have provided correlations between low zinc, carbonic anhydrase activity and protein level.[1803] Zinc supplied to a deficient citrus plant increased RNA and protein synthesis but decreased ribonuclease activity in the leaves.[838] Subsequently, White et al.[1764] reported that RNA synthesis was a prerequisite for protein synthesis. Zinc supplied to deficient Neurospora increased the activity of alcohol dehydrogenase only in the presence of a nitrogen source; apparently protein synthesis must occur to affect enzyme activity. The presence of a nitrogen source appears to be a general requirement for the recovery of zinc-sensitive functions.[1294] When zinc is insufficient in growing organisms, metabolic lesion occurs. First, RNA fails to form, followed by protein, total nitrogen and DNA lesions.[1433,1710] In Euglena severely deficient in zinc, the absolute amount of RNA decreases.[1294] RNA hydrolysis has been found to increase in citrus leaves lacking zinc.[837,838]

Zinc in plants is known to be closely associated with proteins,[1453,1686] and it is bound to some enzymes.[798] Sibly and Wood[1488] found that zinc was not removed by dialysis against water from plant carbonic anhydrase. Zinc is more concentrated in the protein fraction of the Russett Burbank potato tuber rather than in the tuber as a whole, indicating an association between zinc and protein.[926]

Chelating agents remove or combine with zinc cofactors in enzymes, so that enzymatic activity is lost. In yeast alcohol dehydrogenase, 1, 10-phenanthroline does not remove zinc from the enzyme but forms a dissociable zinc-protein-chelate complex that inhibits enzyme activity. This reaction is common to many other zinc metalloenzymes dependent upon pyridine.[1653,1658]

Zinc is so firmly bound in carboxypeptidase that it is not removed by prolonged dialysis against water; however, 1,10-phenanthroline will remove the zinc. Carbonic anhydrase also binds zinc so firmly that its zinc will not exchange with zinc-65 for over 32 days.[712]

There are two types of interaction between zinc and proteins.[928] Zinc metalloenzymes are enzymes in which zinc atoms are specifically and firmly incorporated into the protein, so that they can be considered as a single physical entity in their native state, and homogeneous metalloenzymes can be isolated and identified. In contrast, zinc metal-protein complexes are formed with enzymes which may require zinc as one of several units for activity. The enzymes are more weakly bonded with zinc and cannot be isolated.[928] Less zinc is bound by enzymes or whole plant extracts as the pH of the medium decreases.[798]

Price *et al.*[1296] stated that if zinc were essential to cytoplasmic ribosome stability, then it would be necessary to provide another function of zinc in plant growth and development. Such a function was proposed when it was found that cytoplasmic ribosomes of *Euglena gracilis* contained large amounts of zinc and that the ribosomes became unstable when the *Euglena* lacked zinc.[1289]

Early research showed that zinc and auxin contents are linked in higher plants[1491] and Tsui[1630] concluded that zinc was essential for the synthesis of tryptophan in tomato and indirectly for its auxin synthesis. Later, Nason[1105] found that zinc was needed to form tryptophan from indole and serine.

Because zinc is primarily an enzyme cofactor, it is likely that zinc aids auxin synthesis by activating enzymes.[1453] For example, growth was stimulated in zinc-deficient corn seedlings by adding tryptophan.[1394] Therefore, zinc may be needed to synthesize tryptophan rather than in the direct formation of auxins. The role of zinc in auxin formation is certainly not clear at present. Research also has been reported which postulates that zinc may be involved with starch metabolism in bean plants.[804,805] Again, the research is not yet sufficiently advanced to evaluate this newly proposed function for zinc.

CHAPTER 6

ZINC IN AQUATIC ANIMALS

Zinc is distributed throughout freshwater and marine aquatic environments and occurs in all organisms analyzed. Interest in the accumulation and distribution of zinc in aquatic organisms has arisen from a concern about heavy metals or radioisotopes in human food products of marine or freshwater origin. However, in terrestrial organisms, interest always has been sparked by the need to know nutritional requirements for domestic stock and fowl as well as humans. Therefore, much of the information on zinc in aquatic environments is found in the literature on radioecology and pollution rather than in biochemistry and nutrition reviews. Literature about the ecologic cycling of zinc in aquatic environments and distribution of zinc in aquatic organisms has been written by Bachman,[62] Rice,[1339] Vinogradov,[1695] Bernhard and Zattera,[105] Pentreath,[1223] and Wolfe.[1797,1800]

CONCENTRATIONS OF ZINC IN AQUATIC ANIMALS

Marine Animals

Zinc is present in significant concentrations in all marine animals. The range of concentration among species is relatively narrow, except for oysters, in which very high amounts may occur. Whereas the zinc content of most seafoods ranges from 3-30 ppm, oysters contain 100-2,000^{+} ppm.

The National Marine Fisheries Service has conducted extensive investigations into the content of zinc in numerous species of marine fish, mollusks and crustaceans. These results are summarized in Table 6-1. The average zinc content of all finfish was about 6.5 ppm, and means for individual

TABLE 6-1

Zinc Content of Fish and Shellfish

from Coastal Waters of North America[a]

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
FISH						
Albacore (Thunnus alalunga)	C	8	4.05	1.15	2.00	5.67
Albacore (Thunnus alalunga)	W	10	6.90	9.93	2.64	35.00
Anchovy, Northern (Engraulis mordax)	C	30	23.06	5.39	15.55	44.38
Bass, Black sea (Centropristes striatus)	S	24	4.12	1.70	1.43	9.64
Bass, Striped (Roccus saxatilis)	C	17	3.70	1.06	2.14	6.79
Bluefish (Pomatomus saltatrix)	A	19	9.04	4.09	2.94	19.28
Bluefish (Pomatomus saltatrix)	S	21	8.14	10.78	3.06	54.95
Bocaccio (Sebastodes paucispinis)	C	21	3.86	0.75	2.21	5.00
Bocaccio (Sebastodes paucispinis)	W	21	4.04	0.91	2.14	5.70
Bonito, Pacific (Sarda chiliensis)	C	26	6.67	1.43	2.06	8.57
Butterfish (Poronatus triacanthus)	A	29	7.35	2.30	3.87	14.28
Catfish, Channel (Ictalurus punctatus)	S	10	6.33	2.76	4.42	13.93
Catfish, Gaff-topsail (Bagre marinus)	G	19	10.13	3.75	4.44	17.32
Catfish, Sea (Arius felis)	G	30	14.06	6.33	4.94	30.37
Cod, Pacific (Gadus macrocephalus)	W	37	4.00	1.09	2.50	7.14

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
Cod, Atlantic (Gadus morrhua)	A	46	3.71	1.74	0.87	10.50
Croaker, Atlantic (Micropogon undulatus)	G	10	3.94	0.98	2.50	5.59
Croaker, Atlantic (Micropogon undulatus)	S	10	8.14	11.00	3.20	39.30
Dogfish, Smooth (Mustelus canis)	A	11	4.37	0.93	2.94	6.07
Dogfish, Spiny (Squalus acanthias)	W	44	3.67	1.54	1.61	8.50
Dolphin (Coryphaena hippurus)	H	21	3.46	1.04	1.31	5.53
Dolphin (Coryphaena hippurus)	S	21	5.60	1.27	3.23	8.44
Drum, Black (Pogonias cromis)	G	23	4.81	1.44	20.6	7.50
Drum, Red (Sciaenops ocellata)	G	32	4.08	0.94	2.24	6.43
Eel. American (Anguilla rostrata)	A	10	17.14	4.02	8.75	21.89
Flounder, Gulf (Paralichthys albigutta)	G	20	3.34	0.90	1.40	5.51
Flounder, Summer (Paralichthys dentatus)	S	32	4.83	1.12	2.68	7.51
Flounder, Winter (Pseudopleuronectes americanus)	A	22	4.68	2.23	0.69	12.50
Flounder, Yellowtail (Limanda ferruginea)	A	29	4.73	2.00	1.44	13.06

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
Grouper, Red (Epinephelus morio)	G	10	3.93	0.86	2.50	5.12
Grouper, Black (Mycteroperca bonaci)	G	18	4.85	1.50	3.10	8.31
Grouper, Scamp (Mycteroperca phenax)	S	14	5.18	5.94	1.87	25.51
Haddock (Melanogrammus aeglefinus)	A	32	3.99	3.15	0.13	18.50
Hake, Pacific (Merluccius productus)	W	41	5.08	6.89	2.21	47.10
Hake, Red (Urophysis chuss)	A	19	2.97	1.15	0.40	5.62
Hake, Silver (Merluccius bilinearis)	A	17	3.47	0.64	2.44	4.64
Hake, White (Urophysis tenuis)	A	26	3.01	0.91	1.21	5.44
Halibut, Pacific (Hippoglossus stenolepis)	W	26	3.67	1.47	1.31	7.50
Herring, Pacific (Clupea harengus pallasi)	K	9	14.32	3.92	8.13	22.50
Herring, Pacific (Clupea harengus pallasi)	W	14	5.38	1.78	3.21	8.12
Jack, Crevalle (Caranx hippos)	G	36	6.73	8.66	0.48	56.30
Jacksmelt (whole) (Atherinopsis californiensis)	W	31	12.60	4.12	4.63	25.00
Lingcod (Ophiodon elongatus)	W	49	4.74	1.36	1.54	9.19

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
Mackerel, Atlantic (Scomber scombrus)	A	8	6.84	1.85	4.50	9.42
Mackerel, King (Scomberomorus cavalla)	G	22	5.15	1.27	3.31	8.03
Mackerel, King (Scomberomorus cavalla)	S	20	5.11	1.13	2.25	6.78
Mackerel, Chub (Scomber japonicus)	C	12	7.47	2.56	4.25	14.25
Mackerel, Spanish (Scomberomorus maculatus)	S	29	5.78	1.51	3.82	9.64
Marlin, Blue (Makaira nigricans)	S	11	8.30	3.15	4.00	13.57
Marlin, White (Makaira albida)	S	15	5.05	1.92	2.56	8.57
Menhaden, Atlantic (whole) (Brevoortia tyrannus)	A	23	18.38	5.52	7.75	30.35
Menhaden, Atlantic (whole) (Brevoortia tyrannus)	S	9	15.15	5.50	9.25	23.43
Menhaden, Gulf (whole) (Brevoortia patronus)	G	17	23.85	10.70	11.88	57.14
Menhaden, Gulf (Brevoortia patronus)	G	11	12.67	10.29	5.75	33.93
Menhaden, Yellowfin (whole) (Brevoortia smithi)	G	12	17.17	3.51	13.13	25.00
Mullet, Striped (Mugil cephalus)	G	36	5.56	1.33	2.80	9.64
Mullet, Striped (Mugil cephalus)	S	20	5.95	2.33	3.45	10.75

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
Perch, Ocean (Sebastes marinus)	A	16	3.80	2.40	1.82	12.10
Perch, Silver (Bairdiella chrysura)	S	16	12.19	21.74	4.82	93.60
Perch, White (Morone americana)	A	9	12.28	3.66	6.62	17.90
Pollock (Pollachius virens)	A	22	4.16	2.32	1.12	11.20
Pompano, Florida (Trachinotus carolinus)	G	9	6.50	1.03	5.54	8.81
Pompano, Florida (Trachinotus carolinus)	S	15	8.60	5.64	1.43	26.64
Rockfish, Canary (Sebastodes pinniger)	C	10	6.10	2.92	3.37	10.71
Rockfish, Canary (Sebastodes pinniger)	W	32	4.34	1.21	2.50	8.13
Rockfish, Yellowtail (Sebastodes flavidus)	W	19	4.30	1.59	2.29	8.57
Sablefish (Anoplopoma fimbria)	C	39	3.19	1.65	0.00	9.06
Sablefish (Anoplopoma fimbria)	K	8	2.98	0.80	1.87	4.50
Salmon, Chinook (Oncorhynchus tshawytscha)	K	22	4.83	2.27	2.63	13.75
Salmon, Chinook (Oncorhynchus tshawytscha)	W	55	4.46	1.56	1.38	8.93
Salmon, Chum (Oncorhynchus keta)	K	15	4.66	0.91	2.88	6.34

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
Salmon, Coho (Oncorhynchus kisutch)	K	18	5.52	1.65	3.22	10.00
Salmon, Coho (Oncorhynchus kisutch)	W	25	3.03	0.92	1.30	4.75
Salmon, Pink (Oncorhynchus gorbuscha)	K	15	5.45	1.44	3.57	8.39
Salmon, Sockeye (Oncorhynchus nerka)	K	22	6.24	2.93	2.44	16.07
Salmon, Sockeye (Oncorhynchus nerka)	W	21	7.56	10.10	2.81	49.64
Seatrout, Spotted (Cynoscion nebulosus)	G	28	4.49	1.43	2.34	8.93
Seatrout, Spotted (Cynoscion nebulosus)	S	29	4.34	1.19	2.85	7.50
Shark, Blacktip (Carcharhinus limbatus)	S	10	4.01	1.75	1.25	8.10
Snapper, Red (Lutjanus campechanus)	G	8	3.87	1.38	2.03	6.43
Snapper, Red (Lutjanus campechanus)	S	16	4.90	2.11	2.50	9.64
Snapper, Red (Ehu) (Etelis marchi)	H	9	4.46	0.76	3.28	5.71
Snapper, Vermilion (Rhomboplites aurorubens)	S	25	3.30	0.93	2.06	6.43
Sole, Dover (Microstomus pacificus)	C	28	3.92	1.34	2.14	8.00
Sole, Dover (Microstomus pacificus)	W	38	3.99	1.67	1.18	9.29

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm			
			Mean	Standard Deviation	Low	High
Sole, English (Parophrys vetulus)	C	24	4.59	1.53	1.44	9.19
Sole, English (Parophrys vetulus)	W	40	4.42	1.06	2.38	6.22
Sole, Petrale (Eopsetta jordani)	C	18	3.60	0.87	2.13	5.88
Sole, Petrale (Eopsetta jordani)	W	36	4.32	0.96	2.86	6.61
Sole, Rex (Glyptocephalus zachirus)	C	10	3.89	2.36	1.44	9.82
Sole, Rex (Glyptocephalus zachirus)	W	37	3.63	1.27	1.79	7.00
Spot (Leiostomus xanthurus)	S	19	4.99	0.91	3.21	6.43
Sturgeon, Green (Acipenser medirostris)	W	10	4.33	1.36	2.86	7.06
Tuna, Bigeye (Thunnus obesus)	H	16	3.29	1.07	0.50	4.69
Tuna, Bluefin (Thunnus thynnus)	C	10	6.92	2.87	3.13	14.28
Tuna, Skipjack (Euthynnus pelamis)	C	13	10.39	3.77	4.31	15.55
Tuna, Skipjack (Euthynnus pelamis)	H	16	4.82	1.72	2.13	7.31
Tuna, Yellowfin (Thunnus albacares)	C	22	6.41	2.21	2.40	13.19
Tuna, Yellowfin (Thunnus albacares)	H	15	3.44	1.34	2.31	6.78
Weakfish (Cynoscion regalis)	A	18	4.67	1.96	2.19	10.36

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm Mean	Standard Deviation	Low	High
Weakfish (Cynoscion regalis)	S	10	5.23	1.16	3.75	7.78
MOLLUSKS						
Abalone, Green (Haliotis fulgens)	C	8	18.11	4.86	10.00	26.07
Abalone, Red (Haliotis rufescens)	C	10	8.11	2.56	4.14	13.93
Clam, Butter (Saxidomus giganteus)	W	27	8.04	5.71	0.78	15.72
Clam, Hard (Mercenaria mercenaria)	A	37	22.53	14.44	7.00	58.93
Clam, Hard (Mercenaria mercenaria)	S	9	9.37	1.80	6.56	13.37
Clam, Razor (Siliqua patula)	K	9	10.76	1.65	9.37	14.32
Clam, Razor (Siliqua patula)	W	26	11.88	9.39	0.85	26.92
Oyster, Eastern (Crassostrea virginica)	A	23	271.08	176.32	38.13	1046.88
Oyster, Eastern (Crassostrea virginica)	G	36	156.87	129.33	21.28	455.00
Oyster, Eastern (Crassostrea virginica)	S	31	250.29	68.81	9.81	682.14
Oyster, Pacific (Crassostrea gigas)	C	19	100.90	34.00	48.12	175.00
Oyster, Pacific (Crassostrea gigas)	W	37	228.72	126.64	31.24	912.50
Scallop, Calico (Argopecten gibbus)	S	20	7.09	5.82	0.71	17.14

TABLE 6-1
(Continued)

Species	Area[b]	Samples analyzed[c]	Zinc, ppm Mean	Standard Deviation	Low	High
Squid, Pacific (Loligo opalescens)	C	21	6.98	5.84	1.33	18.13
Squid, Short-finned (Illex illecebrosus)	A	15	16.41	4.91	8.06	35.71
CRUSTACEANS						
Crab, Blue (Callinectes sapidus)	G	15	34.99	32.19	12.50	115.62
Crab, Blue (Callinectes sapidus)	S	11	34.76	8.95	23.00	50.00
Crab, Dungeness (Cancer magister)	W	28	47.98	12.55	24.38	68.57
Crab, King (Paralithodes camtschaticus)	K	10	57.24	38.09	16.31	118.70
Lobster, American (Homarus americanus)	A	16	22.54	6.76	8.81	35.65
Lobster, Spiny (Panulirus argus)	S	18	21.99	16.43	4.69	83.75
Shrimp, Brown (Penaeus aztecus)	G	27	12.63	4.85	4.93	31.07
Shrimp, Pink (Penaeus duorarum)	G	15	9.46	1.34	7.36	11.50
Shrimp, White (Penaeus setiferus)	G	19	6.77	5.93	0.83	20.62
Shrimp, White (Penaeus setiferus)	S	29	8.07	4.91	0.50	15.00

[a]Data from 1975 microconstituent resource survey, National Marine Fisheries Service, College Park, Md. (unpublished).

[b]Areas: A = Atlantic Coast, Newfoundland to Chesapeake Bay
S = Atlantic Coast, Cape Hatteras to Florida
G = Gulf of Mexico
C = Pacific Coast, California
W = Pacific Coast, British Columbia to Oregon
K = Pacific Coast, Alaska
H = Hawaiian coastal waters

[c]All samples are raw edible flesh from individual fish, except when noted.

species ranged from 3-24 ppm. As mentioned, oysters are exceptionally high in zinc among mollusks; their mean for values shown in Table 6-1 is 202 ppm, as compared to a mean value of 12.5 ppm for clams. Crustaceans contained slightly more zinc than finfish; the mean value for crustaceans was 12.5 ppm, and means for individual species ranged from 7-57 ppm.

Such results are consistent with measurements of the concentrations of zinc in shellfish from coastal areas of Scotland,[1626] and amounts of zinc found in finfish sampled from coastal and deepwater areas around England and Wales.[1266] The evidence indicates that fish regulate the concentrations of zinc in muscle tissues and that zinc levels in adult finfish do not vary greatly with age.

Results are also similar for amounts of zinc reported for processed fish and crustaceans. For example, Gormican found the following zinc concentrations in processed samples: canned and salted crab, 36 ppm; uncooked frozen haddock, 3 ppm; canned sockeye salmon, 11 ppm; canned and salted shrimp, 19 ppm; uncooked, frozen sole, 3 ppm; and water-packed canned tuna, 4 ppm.[549] However, Takino reported that the average zinc content in the muscle of 34 species of fish was only about 1 ppm, with a range of zero to 2 ppm.[1591] These values are about tenfold lower than the others reported in previous literature on finfish. The low values and very narrow ranges makes the validity of Takino's research suspect.

Mollusks. The subject of most investigations on zinc in marine animals has been mollusks. Mollusks are able to concentrate certain trace metals up to many thousands of times that level found in the environment.[1301] Pringle et al. have published extensive data on the uptake of zinc and other trace elements by several estuarine mollusks. They collected samples of shellfish at 100

stations from Atlantic and Pacific waters and reported the following zinc contents: Eastern oysters accumulated about 1,428 ppm zinc on a wet weight basis, with a range between 180-4,120 ppm; amounts reported for the Pacific oyster ranged between 86-344 ppm wet weight. The mean zinc content of soft-shell clams was 17 ppm wet weight, and the range was 9-28 ppm. The northern quahog had a mean of 21 ppm wet weight and a range of 12-40 ppm.

Zinc levels in shellfish along the Atlantic Coast varied from about 10-40 ppm in the case of hard- and soft-shell clams to a range between 180-4,100 ppm for the Eastern oyster.[1301] But zinc levels in the Pacific oyster only reached 90-350 ppm. The high levels of zinc found in Eastern oysters suggests a possible genetic difference between Atlantic and Pacific oysters for the physiologic role of zinc.[1301]

Studies on the uptake and content of zinc in soft shell clams showed that clams reached concentrations of 27 ppm in 50 days when exposed to 0.2 ppm zinc at 20 C.[1487] This uptake represented a 17 ppm increase in zinc over a period of 50 days, or 0.35 ppm zinc/kg/day. It was also found that the soft-shell clam apparently concentrates copper preferentially over zinc under identical experimental conditions.[1487]

The significance of chemically polluted estuarine waters for shellfish and, by implication, public health, prompted Shuster and Pringle[1487] to obtain comprehensive data on the levels of certain chemicals in shellfish and study the uptake of trace elements by oysters. Extensive data on zinc in the Eastern oyster (Crassostrea virginica) were compiled; they are summarized in Tables 6-2 and 6-3.

TABLE 6-2

Zinc Content in American Oysters from Atlantic Coast Waters

Source of Data	*Crassostrea virginica* Zinc Content, ppm wet weight	
	Range	Mean
Pringle *et al.*[1301] (Maine through North Carolina, 1965-1967)	204-4,120	1,404
McFarren *et al.*[1009a] (New Hampshire through North Carolina, 1960)	310-4,000	1,641
Galtsoff[522] (Long Island Sound, 1933-1935)	710-2,760	1,468
Chipman *et al.*[275] (Connecticut through Georgia)	740-1,332	1,018

Shuster and Pringle[1487] also studied oysters exposed to 0.1 and 0.2 ppm zinc and produced the data set forth in Table 6-3. Zinc accumulation increased with time over the 20-wk period, and oysters exposed to 0.2 ppm exhibited somewhat higher concentrations than those exposed to 0.1 ppm zinc. The highest levels observed in oysters exposed to 0.2 ppm zinc, however, were comparable to the naturally high concentrations noted in Table 6-3.

TABLE 6-3

Zinc Accumulated by the Eastern Oyster

in Two 20-Week Periods[a]

Week[b]	Crassostrea virginica			
	mean levels zinc, ppm wet weight			
	0.1 ppm exposure		0.2 ppm exposure	
	1967	1968	1967	1968
0	1,036	1,708	1,036	1,708
1	1,456	2,065	1,381	2,265
2	1,561	2,186	1,470	1,856
3	1,538	1,936	1,519	2,030
4	1,831	2,095	1,761	1,474
5	1,956	2,293	2,234	2,267
6	1,736	1,996	1,811	2,200
7	1,666	1,613	2,055	2,451
8	1,732	2,229	1,869	2,307
9	1,911	2,135	2,037	2,343
10	1,570	1,877	1,767	2,531
11	2,035	1,918	2,269	2,413
12	2,189	2,251	2,660	2,642
13	2,059	2,366	2,733	2,222
14	2,139	2,229	2,445	3,051
15	2,212	2,859	2,740	2,912
16	2,118	2,475	2,667	3,233
17	2,382	2,224	3,033	2,869
18	1,970	3,314	3,528	3,159
19	2,206	2,340	2,976	3,743
20	2,708	2,560	3,813	3,185

[a] Data from Shuster and Pringle.[1487]

[b] The mean levels of zinc in the oyster tissue are shown for each weekly sampling interval at two different zinc levels in experimental seawater systems. The experiment, first performed in 1967, was repeated in 1968; trends were similar, although the initial zinc content of the oysters was higher in 1968. The sample was about 200 oysters harvested from Narragansett Bay, Rhode Island.

Oysters lose zinc rapidly if they are transplanted into water with low zinc content.[768] A linear decrease from about 1,000 ppm to about 100 ppm over a 4-mo period was observed. Copper in oysters did not decrease until at least 2 wks after transplanting, whereas zinc started to disappear immediately after the change.

Huggett et al.[745] established a high positive correlation between levels of zinc and levels of copper and cadmium in oysters from the Chesapeake Bay. Based on average values for these elements, they showed that a concentration gradient existed in oysters from all river systems emptying into the bay, and that in the oysters, each metal increased in concentration with proximity to fresh water. The concentration of heavy metals by oysters was shown to follow a predictable pattern: therefore, the oyster is usable as an index for measuring and identifying unnatural amounts of metal taken up.

Although the high content of zinc in oysters has been well documented, little information is available on the biologic availability of zinc in oysters to consumers of seafood. Shah et al.[1478] reported that when rats were fed oyster-supplemented diets containing 60 µg zinc/g diet, the apparent absorption of zinc from oysters was 16.3%, an amount not significantly different from the corresponding value of 18.5% for zinc carbonate ingested by rats. Dietary levels of zinc up to 2,000 ppm from either source had no significant effect on body weight, hemoglobin, hematocrit and liver cytochrome oxidase activity after 8 wks. The concentration of zinc in various tissues was not affected by the source of zinc; however, the dietary level markedly affected the concentration of zinc in all tissues examined

except muscle and hair. A homeostatic control of the intestinal absorption of zinc was demonstrated when the fraction of dietary zinc deposited in rat femurs was found to decrease markedly with increasing dietary concentrations.[1478] The homeostatic mechanism that regulated absorption of zinc appeared to be overcome at a dietary level between 275-1,550 µg/g because the skeletal load of zinc increased at a dietary level of 1,550 µg/g but not at 275 µg/g or less.[1478]

Crustaceans. The zinc content of crustaceans has been studied less than that of mollusks. The zinc content of several whole decapod crustaceans is rather consistent and ranged from 20-50 µg/g (Table 6-4).[186] The overall similarity among the values from different animals suggested that the concentrations of zinc and copper are probably regulated in all decapods. In Homarus, Carcinus, Cancer, and Maja, zinc is found mainly in the blood plasma, rather than in the blood cells, where most of it is bound to proteins in the plasma.[189] The concentration of zinc in the blood is regulated at different values in different species of crustaceans, as listed in Table 6-5. Variations between individuals of the same species appear to be related to the amount of protein in the blood. As Table 6-5 shows, wide differences exist among samples of muscle from different species. Evidence indicates that the zinc content of a muscle may be related to the speed with which it can contract. For example, the muscles that contract quickly in Homarus contain about 15 µg/g zinc, whereas muscles that contract slowly contain about 100 µg/g.[191]

An indication of the permeability of slow and fast muscles in crustaceans was obtained by measuring concentrations of zinc-65 in lobsters kept in sea water to which the isotope was added.[186] The concentrations of zinc-65 (expressed as muscle:blood ratios) in slow muscle were about twice those of

TABLE 6-4

Mean Concentrations of Zinc and Copper in Whole Decapod Crustaceans[a]

Species	Number of Animals	Solid Content, %	Concentration, µg/g wet weight tissue	
			Zinc	Copper
Shrimp and Prawns				
Palaemon serratus	6	27.0	21	30
Palaemon squilla	3	29.7	30	31
Palaemonetes varians[b]	3	24.9	20	32
Crangon vulgaris	6	28.6	34	32
Lobsters and Crayfish				
Homarus vulgaris	1	34.9	23	33[c]
Austropotamobius pallipes	2	28.6	24	17
Galathea squamifera	7	32.7	18	29
Porcellana platycheles	3	44.5	54	27
Hermit Crab				
Eupagurus bernhardus	2	30.2	282	25
Crabs				
Corystes cassivelaunus	1	36.8	39	22
Atelecyclus septemdentatus	3	39.6	32	10
Cancer pagurus	3	33.5	27	20
Portunus puber	3	33.9	27	21
Portunus depurator	3	31.6	21	18
Carcinus maenas	2	28.6	22	22
Xantho incisus	1	34.6	26	20
Pilumnus hirtellus[b]	7	36.6	49	28
Maja squinado	1	33.8	21	25

[a] Data from Bryan.[186]
[b] Females with external eggs.
[c] Mean of 10 animals.

fast muscle, suggesting that slow muscle is more permeable to zinc. The nature of the differences in concentrations of zinc between the two types of muscle is not known. This feature, however, suggests that lobster, and presumably other decapod crustaceans, would be suitable organisms for determining how zinc functions in the muscles of crustaceans.

Concentrations of zinc in the hepatopancreas vary from species to species, but usually lie within the range of 30-90 μg/g (see Table 6-5). In Homarus, dietary zinc is rapidly absorbed and the concentration increases in the blood and hepatopancreas. It is thought that the zinc stored in the hepatopancreas of Homarus is not removed in the feces. Rather, it is lost across the body surface or gradually lost into the blood and then excreted in the urine.[193]

The amount of zinc which can be absorbed directly from seawater across the body surface of decapod crustaceans increases if the concentration in the water is raised. Therefore, species living in fairly clean water will absorb less zinc from the water than species living in a polluted water where the concentration is higher. However, variation in the dietary intake is probably more important.[186]

Freshwater Animals

Data on the content of zinc in freshwater animals are limited, but it seems that the levels in freshwater fish are not markedly different from amounts found in marine animals.

The concentration of zinc and 12 other elements was measured in a limited study of dressed fish from nonindustrialized and heavily industrialized freshwater areas around the Great Lakes.[1648] All samples were composite, consisting of at least 2.25 kg or 3 fish. No major differences were detected between

TABLE 6-5

Mean Concentrations of Zinc in Body Fluids and Tissues of Decapod Crustaceans[a]

Species[b]	Number of Animals	Zinc, µg/g fresh weight					
		Blood	Leg Muscles	Abdominal Muscles	Stomach Fluid	Hepato-pancreas	Urine
Palaemon serratus	6	38	--	10	--	64	--
Palaemonetes varians	6	87	--	14	--	65	--
Crangon vulgaris	6	23	--	14	--	78	--
Palinurus vulgaris	2	3.1	66	20	48	97	0.6
Homarus vulgaris	4	7.4	60	15	0.7	34	2.2
Austropotamobius pallipes pallipes	6	0.9	--	12	41	109	0.02
Galathea squamifera	5	0.25	--	10	47	49	--
Eupagurus bernhardus	3	11.6	36	24	26	69	--
Corystes cassivelaunus	5	11.0	52	--	12	50	--
Atelecyclus septemdentatus	3	9.5	61	--	6	88	1.4
Cancer pagurus	5	49	64	--	15	45	0.6
Portunus puber	5	7	28	--	15	42	0.2
Portunus depurator	3	1.8	15	--	13	24	0.3
Carcinus maenas	11	36	44	--	18	56	0.4
Pilummus hirtellus	3	12.3	--	--	92	169	--
Maja squinado	3	2.4	63	--	31	71	0.3

TABLE 6-5 (continued)

Species[b]	Number of Animals	Zinc, μg/g fresh weight: Excretory Organs	Gills	Shell	Vas Deferens	Ovary	External Eggs
Palaemon serratus	6	--	--	--	--	--	24
Palaemonetes varians	6	--	--	--	--	--	--
Crangon vulgaris	6	--	--	--	--	--	--
Palinurus vulgaris	2	20	20	16	--	82	107
Homarus vulgaris	4	20	15	5	13	50	--
Austropotamobius pallipes pallipes	6	7	8	8	14	26	--
Galathea squamifera	5	--	27	9	--	--	--
Eupagurus bernhardus	3	23	69	28[c]	27	56	--
Corystes cassivelaunus	5	16	45	5	27	--	--
Atelecyclus septemdentatus	3	29	27	7	30	--	--
Cancer pagurus	5	29	42	3	16	--	--
Portunus puber	5	10	23	13	20	87	--
Portunus depurator	3	8	25	3	23	--	--
Carcinus maenas	11	19	26	3	23	--	--
Pilumnus hirtellus	3	--	60	17	--	--	--
Maja squinado	3	15	10	5	16	45	--

[a]Data from Bryan.[186] [b]Samples are from English waters. [c]Shell not heavily calcified.

the zinc levels of fish from nonindustralized and industralized areas. Lake whitefish and northern pike from the nonindustralized Moose Lake area had 14 and 19 ppm zinc, respectively. Their counterparts in parts of Lake Ontario and Lake St. Pierre affected by industry had incorporated 12 ppm and 19 ppm zinc. Northern pike, rainbow smelt, and yellow perch around industralized areas of Lake Erie had absorbed 10, 20, and 12 ppm zinc, respectively.[1648]

Lucas et al. also investigated the concentration of zinc and other trace elements in fish from the Great Lakes.[954] They studied the content of zinc and 14 other trace elements in 40 liver samples from 10 species of fish from Lakes Michigan, Superior, and Erie. The levels of zinc varied little between species and the range was 11-48 ppm. This range is somewhat higher than the range (3-24 ppm) reported in Table 6-1 for mean concentrations of zinc in marine samples.

Concentration of Radioactive Zinc

Levels of zinc-65 have been studied in marine organisms mainly for their relation to discharges from nuclear reactors. Pearcy and Osterberg[1215] investigated the major γ-emitting radioisotopes found in the livers of albacore (Thunnus alalunga) along the west coast of North America from 1962-1965. The zinc-65 content of albacore livers from southern California and Baja California ranged from 10-100 pCi/g, which was about 10% of the levels found in samples off Oregon and Washington.

The highest levels of zinc-65 were found in samples taken off northern Oregon and Washington, and those levels increased markedly during the summer. The data suggest that albacore rapidly accumulate zinc-65 after they migrate into Oregon waters. It was concluded that radioactivity from the Columbia River significantly affects the zinc-65 content of migratory albacore tuna. Zinc-65 content and specific activity in albacore tended to be higher in fish taken nearer the mouth of the Columbia River and in those taken in the summer and fall months.[1215]

The levels of zinc-65 in benthic invertebrates of the Oregon coast have been reviewed by Carey.[231] Zinc-65 and other artificial radioisotopes in the marine environment off the northwest coast of the United States come from activation of naturally occurring stable elements and from corrosion of reactor parts within the Atomic Energy Commission (AEC) reactor at Hanford, Washington. Marine benthic fauna concentrate zinc-65 and the concentration changes markedly with depth.[232] Rapid decreases generally occur within the first 400 m of depth.

Preston[1290] reported that the rate of discharge of zinc-65 by a nuclear power station in Essex, England, is based on the amount of the metal that accumulates in oysters in the surrounding area. The rate of discharge of zinc-65 is restricted specifically because of its high concentration in oyster flesh. Assuming maximum human consumption of oyster flesh of 75 g/day over the whole year, the calculated permissible level for oyster flesh was calculated to be about 2,900 pCi/g. Because it had been shown that the concentration factors $\frac{\text{concentration of zinc-65 accumulated in organism tissue}}{\text{concentration of zinc-65 in seawater}}$ for zinc tended to increase as the level of zinc in the seawater decreased,[275] Preston was able to suggest that the accumulation of zinc-65 in oysters could be limited by adding inactive zinc to seawater.[1290]

Studies on the stable zinc concentration in oysters before opening the Essex power plant reported a mean of 367 ppm, with a range of 308-419 ppm, whereas when the power plant was operating the mean was 834 ppm, with a range of 388-1,230 ppm. These data indicated that zinc levels had risen, perhaps caused by the zinc in the pipework of the power station's condenser cooling water system.

ZINC METABOLISM IN AQUATIC ANIMALS

Mollusks

Oysters. The accumulation of zinc by various members of the family Ostreidae received attention over half a century ago,[136,704] but that interest was generated primarily by the undesirable phenomenon of "greening" in commercial oysters, the outcome of excessive tissue concentrations of metallic elements, particularly copper.[521,522] More recently, the accumulation by oysters of zinc-65 from nuclear fallout[475,1097,1797] or reactor effluents[1290,1397,1477,1738] renewed interest in studying zinc metabolism in oysters and other organisms. It has been found that oysters may concentrate zinc to levels greater than 100,000 times the ambient concentrations in seawater,[275,1290,1797] and an inverse relationship was demonstrated between this concentration factor and the zinc concentration in seawater, suggesting a limiting or regulatory process.[275,1290] This mechanism may exist because of the limited number of zinc-binding sites on structural proteins.[1223] Curiously, replotting the data reported reveals a strong linear log-log correlation between "equilibrium zinc concentrations" in oysters and seawater, a correlation that might be expected from a simple adsorption mechanism.[275,1290]

Zinc is not strongly localized in particular organs or tissues of oysters;[312,768,1797] nor are there great differences in the molar ratios of calcium, magnesium, sodium or potassium compared to zinc in any tissues.[312] External tissues such as mantle, gills, and labial palps tend to contain more zinc than do internal tissues; adductor muscle tissue usually contains

the least. In all tissues, zinc commonly is found with nuclear and cell debris and with cell sap or cytoplasm, rather than being localized in either microsomal or mitochondrial fractions.[312,1361,1800] However, in none of these studies was the relative concentration of zinc determined in any of the subcellular fractions.

Nearly all soluble zinc in supernatant preparations from homogenized oyster tissues is bound to proteins of high molecular weight, as experiments with gel-diffusion chromatography have shown.[1361,1800] Dialysis of whole oyster tissue homogenates[312,1361] or soluble extracts,[1800] however, removes about 95% of the zinc. Therefore, the zinc may be very weakly bound and readily dissociated from the protein moieties. Based on the observation that removing 95% of the zinc present had no effect on the activity of alkaline phosphatase, Wolfe[1800] concluded that most of the zinc in oysters was superfluous to the biochemical or nutritional requirements of the organism. Similarly, calculations of the zinc requirements of the oyster based on enzyme-specific activities and zinc stoichiometry recorded for zinc enzymes purified from other biologic systems amount to a small fraction (about 0.1%) of the total zinc present in oysters.[312,1224] Of the enzymes demonstrated to be zinc metalloenzymes in other systems, only four (carbonic anhydrase, alkaline phosphatase, carboxypeptidase A, and malic dehydrogenase) have been detected in Ostrea edulis[312] or Crassostrea virginica.[1800] Alpha-D-Mannosidase, a zinc metalloenzyme previously described for a gastropod mollusk (the limpet),[1520] was also abundant in Ostrea edulis.[312] The zinc metalloenzymes alcohol dehydrogenase, lactic dehydrogenase, glutamic dehydrogenase, and carboxypeptidase B were not detected in whole tissue homogenates or subcellular fractions from either species of oyster.

Two hypotheses--both related to the metabolism of calcium--have been offered to explain the high zinc concentrations found in oysters. The first postulates that zinc may be assimilated from the environment coincidentally with calcium by a relatively nonspecific ion transport mechanism to satisfy the organism's large calcium requirements for shell deposition.[1800] Moreover, at the mantle and shell interface, effective ionic discrimination occurs, such that calcium is deposited in the shell and zinc remains loosely bound to available sites in the soft tissues. According to the second hypothesis, zinc would have to be accumulated selectively and actively to overcome the competitive action of calcium and maintain the vital functions dependent upon zinc, especially the activities of carbonic anhydrase and alkaline phosphatase, which are necessary for shell formation.[312]

This possible zinc-calcium interaction also may be studied by considering research on the dependency of relative ion composition of oyster tissues on salinity and relative turnover rates of zinc and calcium.[745,746] Because calcium content of seawater is directly correlated with salinity and zinc content is not, significant differences in zinc concentration or turnover as a function of salinity would be expected for either of the mechanisms proposed above.

To satisfy the first hypothesis, the zinc concentration would have to increase at reduced salinities, because the calcium-assimilative transport system presumably would have to work harder to maintain sufficient calcium reserves for shell deposition. Conversely, if the second hypothesis is correct, reduced zinc turnover and concentration in oysters at lower salinities would be expected because of the decreased competition from environmental

calcium. To date, only one experimental study has been conducted in which the effects of salinity were separated from the influences of other significant environmental variables. Duke et al.[406] studied zinc-65 accumulation by Crassostrea virginica and other estuarine organisms in a multifactorial experiment in which salinity, stable zinc, pH and temperature were each tested at two different levels for accumulation over 15 days.

The effect of salinity, tested at 25 and 30 ppt, was significantly inverse: that is, zinc-65 accumulation was about 10% lower at the higher salinity. An inverse salinity effect was also noted on the contents of manganese and magnesium plus strontium in shells,[1383] implying that the oyster accepts less preferred ions for shell construction when subjected to lower environmental calcium concentrations.

Copper and zinc were analyzed in oysters (Crassostrea virginica) from various stations in the Newport River in North Carolina, and the Rappahannock River in Virginia. Higher concentrations of copper and zinc were found in animals living in fresher waters as had been shown previously for oysters in the James, York, and Rappahannock estuaries in Virginia.[746] For example, in the Newport River estuary, oysters living at about 12 ppt salinity contained a mean of 320 ppm zinc wet weight, whereas oysters at 32 ppt contained about 130 ppm zinc. The concentrations of zinc in unfiltered water samples from this same estuary were constant, irrespective of salinity,[339] implying that the concentration gradient found in the oysters does not result from a similar gradient in the water.

The biologic turnover of zinc by oysters and other aquatic organisms is summarized in Table 6-6. The most reliable data on zinc turnover in oysters are from Seymour's studies conducted in the natural environment for two years.[1476] These data, summarized in Table 6-6, were recently analyzed again by Cutshall,[347] who showed that both the accumulation and loss data were described by simple first-order exponentials with similar rate constants. The effective half-life for zinc-65 turnover was 135 days, and the corresponding biologic half-life was 300 days. Thus, once accumulated by oysters, zinc is retained very effectively.

TABLE 6-6

Zinc Turnover in Aquatic Organisms

Species	Biologic Half-Life	Experimental Conditions
Mollusks		
Mytilus edulis[a]	3.3-3.9 days	Up to 20-day zinc-65 uptake; 70-day loss in laboratory; animals unfed
Mytilus californianus[b]	76 ± 3.5 days	Columbia River zinc-65; life-long accumulation, one year of loss in natural environment
Crassostrea gigas[c]	300 days	Columbia River zinc-65; life-long accumulation; 2 years of loss in natural environment
Anodonta nuttalliana[m]	650 days	Single injection of zinc-65; loss to synthetic running pondwater
Littorina irrorata[i]	40 days @ 15 C 25 days @ 25 C 23 days @ 30 C	
Crustacea		
Homarus vulgaris[d]	60-270 days	Unspecified, but turnovers correlated positively with zinc concentration of seawater
Maja squinado[e]	38 days	Injection of zinc-65; loss into seawater containing 100 µg/l zinc; periodic feeding
Austropotamobius pallipes pallipes[f]	30-38 days	Single feeding of zinc-65; loss into unlabeled freshwater; stable zinc in food increases turnover
Euphausia pacifica[g]	140 days	15-day feeding period with zinc-65 labeled *Artemia nauplii*; 5-mo loss period
Anonyx sp.[h]	140 days @ 3 C 100 days @ 7 C 90 days @ 12 C	6-10 day zinc-65; accumulation from seawater; 29-day loss period; data obtained with and without sediment were combined
Fish		
Fundulus heteroclitus[j]	75 days @ 10 C 58 days @ 20 C 35 days @ 30 C	
Pleuronectes platessa[k]	295-313 days	267-day zinc-65; accumulation from seawater; 91-day loss period
Micropogon undulatus[l]	6.5 days	Single intraperitoneal injection of zinc-65; loss to clean seawater

[a] Derived from van Weers.[1674]

[b] Derived from Young and Folson.[1814]

[c] Derived from Seymour.[1476]

[d] Derived from Bryan *et al.*[197]

[e] Derived from Bryan;[186] see also Table 6-5.

[f] Derived from Bryan;[192] see also Table 6-5.

[g] Derived from Fowler *et al.*[502]

[h] Derived from Cross *et al.*[338]

[i] Derived from Mishuma and Odum.[1069]

[j] Derived from Shulman *et al.*[1486]

[k] Derived from Pentreath.[1222]

[l] Derived from Baptist *et al.*[71]

[m] Derived from Harrison.[637]

Marine mussels and scallops. Mytilus has been used extensively as an indicator of radioactive contamination because of its local abundance and worldwide distribution; consequently, considerable attention has been focused on zinc metabolism in this organism. Values for zinc turnover have been obtained for Mytilus edulis L.,[1222,1477,1674] Mytilus californianus,[899,1477,1814] and Mytilus galloprovincialis,[826] and they are listed for the first two species in Table 6-6. Keckes et al.[826] demonstrated conclusively that the experimental determination of short-term turnover rates using zinc-65 and resolution of turnover into differing rate constants depended heavily upon the duration of exposure of the organism to zinc-65. From these observations, it is evident that many mechanisms interact to produce zinc turnover, and that long exposure periods are necessary for valid measures of elemental turnover in radioisotopic loss experiments. This relationship is discussed theoretically by Cutshall;[347] it is evident specifically in comparisons of the data from experimental exposures of various short durations,[826,1674] and in data from lifelong exposure to zinc-65,[1814] which suggested only a single long-lived component when zinc-65 was lost by Mytilus.

Unlike oysters, where zinc is generally found in most tissues, zinc is strongly localized in certain tissues of Mytilus. Tissue concentrations of zinc in Mytilus edulis aoteanus have been reported in the order of visceral mass > gills > gonads > mantle > muscle = foot = shell.[168] Pentreath[1223] found visceral mass > adductor muscle > gonad > mantle = foot = gill. The distribution of zinc-65 fallout was examined in Mytilus californianus,[1814] where half the total zinc-65 was found in the kidney tissues constituting about 1% of the total weigh; an additional 19% of the isotope was discerned in the digestive

glands representing less than 4% of the weight of Mytilus. Only 4% of the zinc-65 was in the shells, which constitute about 70% of the weight. Presumably, the "visceral mass" measurements just noted [168,1222] include kidney tissue. The localization of zinc in the kidneys was confirmed by autoradiography of zinc-65 accumulated experimentally.[1674] However, in a related species of Mytilidae, Modiolus modiolus, rather uniform zinc concentrations were found in gonad, mantle and gills, gut, and digestive gland, and they were 2-2.8 times higher than that in muscle tissue.[1467] Although it is possible that zinc metabolism varies from species to species, it is more likely that greater care should have been exercised during dissection to demonstrate the specific tissue distribution of zinc.

Scallops (Pectenidae) also show strong tissue localizations of metals, including zinc. Again, kidney tissue exhibits highest concentrations of zinc, 10-100 times the concentration for the total soft parts, in Pecten novae-zelandiae,[168] Pecten maximus L., and Chlamys opercularis L.[190] High zinc concentrations in the kidneys usually are accompanied by high concentrations of manganese and iron.[168,190,1422,1467] Some of the data[190] show similar relationships between kidneys and the various other tissues which also hold true for cobalt, copper, nickel, and lead, which are summarized in Table 6-7. Although metal concentrations were usually highest in kidneys, the digestive gland frequently contained a significant portion of the total metal present in the scallop, because the kidney is such a small organ. Seasonal fluctuations were evident in the kidney and digestive gland: zinc and manganese concentrations were maximal in late autumn and winter and minimal during

TABLE 6-7

Relative Content of Metals in Different Scallop Tissues[a]

Metal, %[b]	Pecten maximus L.				Chlamys opercularis L.			
	Gills	Mantle	Digestive Gland	Kidneys	Gills	Mantle	Digestive Gland	Kidneys
Aluminum	--	--	33.9	0.7	--	--	7.4	0.8
Cadmium	--	--	89.9	1.7	--	--	41.5	7.5
Chromium	--	--	46.9	1.7	--	--	18.1	3.0
Cobalt	6.7	8.8	50.3	23.9	11.5	8.7	26.0	36.2
Copper	5.3	4.4	60.9	1.5	7.9	4.6	17.3	57.6
Iron	5.7	7.4	71.9	0.6	9.7	8.4	61.4	2.3
Lead	3.1	5.4	19.0	52.3	2.1	1.8	10.2	78.8
Manganese	3.1	1.2	1.4	92.5	5.7	1.7	1.6	89.0
Nickel	7.2	7.9	45.9	20.0	9.1	6.1	24.5	43.0
Silver	--	--	63.7	1.5	--	--	62.4	3.4
Zinc	3.0	3.6	16.2	51.8	7.4	5.1	2.6	75.1

[a]Data from Bryan.[190]

[b]Percentage of total metal in soft parts, including fluid.

spring, summer and early autumn.[190] Bryan suggested that seasonal variations may be caused by the reproductive cycle, temperature, availability of food, and land drainage; he surmised that elemental changes in the kidney were independent of the reproductive state of the gonads, and that changes in phytoplankton productivity were probably more important than either temperature or runoff. Generally, periods of highest productivity coincided with lowest metal concentrations in scallop tissues. Bryan further reasoned that the greater availability of phytoplanktonic food would increase the metabolic rate of the scallop and hence the excretion of waste products; reduce the amount of metal left available in the water; and diminish the metal concentration per phytoplankton cell, thereby decreasing metal intake of the scallop despite the additional food ingestion.

Freshwater bivalves. Freshwater bivalves are well known for their concentration of manganese,[1032,1695] generally accompanied by a lower concentration of zinc.[637,638,648] As in the scallops, these metals are unevenly distributed among the tissues of unionid bivalves. Anodonta nuttalliana Lea has a mass of tissue in which approximately 25% of the dry weight is composed of calcium. This calcareous tissue contains metal-rich granules 2 μm in size, and is present as an elongated white area on the surface of the mantle near the attachment of the gills.[637] Radionuclides of manganese, cobalt, zinc and lead are rapidly accumulated to high levels in this tissue, whereas accumulated radionuclides of scandium, chromium, iron, europium, tantalum, and mercury were found in the organs of the digestive system.[638] After 147 days of accumulation, the gonad and foot of the clam appeared to have equilibrated with both zinc-65 and manganese-54, but the mantle and calcareous tissue were still accumulating isotopes at an exponential rate.

The biologic half-time for turnover in the large, long-lived pool in Anodonta was about 1,300 days for manganese and 650 days for zinc.[638] Of the total zinc-65 accumulated in 35 days, about 35% was in the calcareous tissue, whereas the proportion of stable zinc in this tissue was 42% in small clams (in which average wet weight of tissue was 15 g), and about 58% in large clams (where wet weight averaged 45 g or more).[637]

In similar studies with Anodonta californiensis, Pauley and Nakatani[1214] found that zinc-65 was associated predominantly with mantle and gill tissue, but they did not distinguish calcareous tissue. Nor did their autoradiographic techniques identify the granules observed by Harrison.[637] Instead the autoradiography showed highest localizations of zinc-65 in the base and tip of the outer mantle epithelium, the tips of the gill epithelial cells, and the epithelial cells of the kidney. It seems probable that Pauley and Nakatani simply included the calcareous tissue either with gills or mantle tissue, and they probably did not section the organism through this region. A similar judgment may be applied to the distribution of zinc-65 in tissue of Margaritifera margaritifera.[1024] In studies of whole animals, rapid equilibration with zinc-65 has been noted for several species of unionids.[183,648,1024] But the slow turnover and high concentrations of zinc in the calcareous tissue,[637] probably thwarted the achievement of true equilibration, and some of the biologic half-lives obtained[648,1024] are probably not representative of zinc turnover in these species.

Freshwater gastropods. In a freshwater prosobranch, Viviparus malleatus, zinc-65 was accumulated to much higher concentrations in soft parts than in the shell, but tissue distribution was not studied.[183] Unborn embryonic snails contained a higher concentration of zinc-65 than did the adults collected at the same time from the experimental pond, suggesting that zinc mobilization into the gonads and internally developing embryos is rapid compared to turnover in some other organs of the adult snail. Stable zinc was not analyzed in this study; therefore, dynamics of zinc turnover were not well defined.[183]

Calcium accumulation and turnover were studied in the pulmonate Lymnaea stagnalis.[1678] Calcium was pumped against a concentration gradient by this gastropod from low concentrations in the water (< 1 mg calcium/l). The calcium transport system was directly affected by temperature; the rate of calcium uptake showed a Q_{10}* of 1.4 between 6 C and 16 C and 3.0 between 16 C and 30 C. At least two rate components were resolved for calcium exchange between Lymnaea and the medium. Although zinc was not measured in this study, it is mentioned here because of the implied relationship in oysters between calcium metabolism and the bioaccumulation of metals like zinc and manganese, which was discussed in the section on oysters.

Crustaceans

Zinc-65 was assimilated with about 56% efficiency from ingested food (zinc-65-labeled Artemia) by the gammarid amphipod Anonyx and retention of the isotope was affected significantly by feeding and deprivation.[338] Amphipods starved during the 3-wk loss period exhibited a mean biologic half-life

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* Increase in rate of chemical reaction for each 10 C increase in temperature.

for zinc of about 100 days, compared to about 35 days for animals on a brine shrimp diet. Similar effects of controlling the food allowance on zinc-65 turnover were observed in marine Idothea[1160] and crabs.[192] Turnover of zinc by Anonyx was also directly related to temperature; biologic half-life for zinc turnover was about 92 days at 12 C and about 150 days at 3 C.[338] Similar effects of temperature were also noted with the isopod Idothea,[1160] with the euphausids Euphausia pacifica and Thysanaessa spinifera,[503] and with the crab Callinectes sapidus.[1339]

Autoradiography of crustacean tissues indicates that zinc-65 is located primarily in the interstitial spaces between muscle fibers, in the eye mainly between the rhabdoms and cystalline cones of adjacent ommatidia, within and on the interior surface of the exoskeleton, regardless of whether the isotope was accumulated directly from water[340,503] or assimilated from labeled food.[501] In euphausids, the prawn Pasiphaea pacifica and shrimp Pandalus tended to concentrate zinc-65 accumulated from water more in the exoskeleton and less in muscle than when the isotope was accumulated from food. In all cases, however, the exoskeletons contained 30-66% of the accumulated zinc-65.[501]

The turnover of zinc by growing crustaceans was not readily studied in short-term zinc-65 loss experiments because a mean of 41% of the body burden of zinc-65 was lost with the molted exoskeletons after zinc-65 accumulation from water.[503] In long-term loss experiments after ingestion of zinc-65-labeled food, however, only 1% of the body burden was excreted with each molted exoskeleton.[502] This finding corresponded to the earlier radioautographic observations that zinc-65 accumulated from water tended to be associated

with interstitial spaces in the calcified cuticle of the exoskeleton, whereas ingested zinc-65 was more localized on the underside of these cuticular layers.[340,501,503]

Fowler et al.[501] speculated that the localization of zinc in euphausid eyes might be associated with melanin pigments in the distal and proximal screening pigment cells that closely surround the cones and rhabdoms of euphausid eyes.[814] Zinc also may be required by the enzyme retinene dehydrogenase, which catalyzes the oxidation of vitamin A alcohol (retinol) to vitamin A aldehyde (retinene). Zinc is necessary to many other nicotinamide adenine dinucleotide (NAD)-dependent enzymes.[1664]

Based on the above mentioned research on zinc-65 accumulation and turnover in euphausids, and studies of zinc-65 accumulation by Euphausia pacifica under conditions simulating those which euphausids would encounter if they migrated vertically in and out of a zinc-65-labeled mixed surface layer of the ocean at different seasons in temperate latitudes,[1499] the turnover of zinc by the single euphausid Meganyctiphanes norvegica[1501] as well as by total populations of this organism in the Mediterranean region[1500] was estimated. Flux of zinc through Meganyctiphanes norvegica was presumed to follow a linear model where:

$$K_{\underline{e}} = \mu_{\underline{e}} + \lambda_{\underline{e}},$$

$$K_{\underline{e}} = Q_{\underline{i}}\, \rho_{\underline{i}},$$

$$\mu_{\underline{e}} = Q_{\delta}\, \rho_{\delta},$$

$$\lambda_{\underline{e}} = Q_{\underline{f}}\, \rho_{\underline{f}} + Q_{\underline{m}}\, \rho_{\underline{m}} + Q_{\underline{c}}\, \rho_{\underline{c}} + Q_{\underline{x}}\, \rho_{\underline{x}}, \text{ and}$$

$Q_{\underline{i}}$, Q_{δ}, $Q_{\underline{f}}$, $Q_{\underline{m}}$, $Q_{\underline{c}}$, and $Q_{\underline{x}}$ are respective zinc concentrations

in ingested food (i), new tissue added in growth (δ), feces (f), molts (m), dead carcasses (c), and nonviable eggs (x); and ρ_i ρ_δ, ρ_f, ρ_m, ρ_c, and ρ_x are the rates of ingestion (i) and growth (δ), and the respective rates of production of feces (f), molts (m), dead carcasses (c), and nonviable eggs (x). For adult animals feeding and defecating 12 h/day, zinc ingestion rate (K_e) was estimated as 51-130 µg zinc ingested/g dry weight/day, depending on whether maximal or minimal values of the other concentrations and rates were used. The zinc concentration in the food of Meganyctiphanes norvegica would have to be 400-450 µg zinc/g dry weight to satisfy the elimination and growth rates of the model. Because zinc measured in Artemia fed to Meganyctiphanes norvegica in the laboratory (417 ± 103 µg zinc/g dry weight) and in natural plankton mixtures consisting of 95% small copepods plus phytoplankton, flagellates, and detritus (570 ± 113 µg zinc/g dry weight) hewed closely to the calculated estimate, the zinc budget for Meganyctiphanes norvegica was believed to be described adequately. Fecal pellet deposition represented over 90% of the total zinc flux from the organism. For the Ligurian Sea, the entire pool of ionic zinc in the water would be circulated through the Meganyctiphanes norvegica population in 500-1,200 yr, depending on the dietary availability of zinc.[1500] Furthermore, zinc would be effectively transported downward from the surface waters by sinking fecal pellets, molts, and carcasses generated by the vertically-migrating euphausid population during daylight hours. Daily net transport of 36-98% of the pool of body zinc in Meganyctiphanes norvegica would occur to a depth of 500 m, depending on whether food availability was marginal or sufficient, and at least 6% of the body zinc pool would reach 2,500 m daily.

Osterberg et al.[1183] attempted to correlate the distribution of several radionuclides including zinc-65 with the exposed surface areas of macroplanktonic organisms, and concluded that surface adsorption played a relatively insignificant role in the bioaccumulation of the nuclides. Yet Fowler et al.[503] found that weight-specific uptake and elimination of zinc-65 were statistically similar between live and formalin-preserved euphausids over a range of temperatures and zinc-65 concentrations. In euphausids which apparently do not regulate zinc concentrations in tissue, the turnover and steady-state concentrations of zinc probably depend entirely on the number and affinities of internal binding sites for the metal.[502] A similar hypothesis also was suggested for zinc concentration and turnover in oysters.[1800]

In large decapod crustacea (crabs, lobsters, and crayfish), zinc concentrations in the body are controlled by mechanisms regulating zinc absorption through the gills and zinc loss in the urine and across the body surface.[186, 191-193]

The following discussion is derived from Bryan's reviews of zinc metabolism in decapods.

The zinc concentration in blood of lobsters (Homarus vulgaris) from waters containing about 5 µg zinc/l is similar to the value of 5.6 µg zinc/g given for human blood.[376] Whereas in human blood about 90% of the zinc is found in the erythrocytes and leukocytes, in lobster blood 93% of the zinc is in the serum. As in vertebrates, this zinc appears to be bound to blood protein, which in the lobster is principally the copper protein, hemocyanin. Normal hematic zinc concentration is highest in lobsters that have more hemocyanin, measured by high contents of copper and solids in the blood.

In normal male lobsters, the highest concentration of zinc is found in the hepatopancreas: about 25 µg/g. High zinc concentrations were not

found to be associated with the male reproductive system in the lobster (the vas deferens contains about 13 μg/g). But the highest zinc concentrations in normal female lobsters were found in the ovary, which contains up to 50 μg/g. No other obvious differences between the sexes were observed. Zinc concentrations in normal lobsters are relatively low compared to vertebrates, but are very similar to the zinc concentrations of 20 μg/g found in whole shrimp and crabs.[136,1209]

Bryan[193] has showed that zinc concentrations in most lobster tissues are quite accurately controlled. Except in the hepatopancreas, variations produced by zinc injections were rectified quite rapidly. In muscle tissue and gonads no changes were induced by zinc injections, suggesting that these organs are either almost impermeable to zinc or that zinc regulation is particularly good.

The process of zinc regulation in the freshwater crayfish (Austropotamobius pallipes) differs from that in the marine lobster Homarus because most crayfish tissues contain less zinc than those of the lobster, but the concentrations in the hepatopancreas and stomach fluid are much higher. Concentrations in the main abdominal flexor muscles are similar in the two species, however, and zinc concentrations in muscle are fairly constant under different conditions. In both species, muscle is the tissue responding least to changes in the blood concentration of zinc, although zinc exchange does occur.[193]

Crayfish blood contains less zinc than the blood of any marine decapod crustaceans that have been examined, although in estuarine species blood zinc was higher than in marine species. The blood concentrations of zinc do not increase in crayfish, despite high concentrations of blood protein and copper which tend to increase amounts of zinc in Homarus. Zinc is bound

to proteins in the blood, but binding to the variable hemocyanin component may be less important for crayfish.

Low zinc concentrations for crayfish have been found consistently in the excretory organs and urine. The excretory system appears to be unimportant in regulating zinc, although it certainly prevents zinc loss. The permeability of the body surface to zinc is low and therefore zinc losses are small. Similarly, very little zinc is absorbed across the body surface from solution. In contrast, Homarus absorbs and loses zinc across the body surface, apparently in a controlled manner. Urinary losses in Homarus are also closely controlled, yet they can be appreciable.[192]

Both species are likely to obtain excess zinc from food, and it is the major source of zinc for the crayfish. Excess zinc is absorbed rapidly from the stomach fluid in Homarus, partly by the hepatopancreas, and concentrations will increase in all tissues except muscle, gonads and exoskeleton. Uptake from the stomach fluid of the crayfish is slower, and all the zinc is absorbed by the hepatopancreas before affecting the other tissues. If zinc penetrates directly from the stomach of the crayfish into the blood, it is removed so rapidly by the hepatopancreas that no obvious change in blood concentration is seen. When excess zinc was injected into crayfish blood, all the excess zinc was absorbed by the hepatopancreas in a few days and some of it was transferred to the stomach fluid. When excess zinc was injected into Homarus, some was removed in the urine, some was lost across the body surface, and as in the crayfish, an appreciable amount was removed by the hepatopancreas. Whereas in the crayfish excess zinc in the hepatopancreas and stomach fluid

was lost in the feces, in Homarus excess zinc in the hepatopancreas was eventually lost by blood, excretory organs, or body surface.[192]

The amount of zinc absorbed directly from sea water across the body surface of decapod crustacea varies with the zinc concentration in the water. Hence species living in fairly clean water away from the coast will absorb less zinc from the water than species living in lightly polluted estuaries. Variation in the dietary intake of these metals is, however, probably much more important. Animals which feed on worms from sediments may take in considerable zinc and copper. Although the intake of metals may be highly variable, the concentrations of zinc and copper in the majority of species lie between 20 and 35 µg/g. Therefore, it seems likely that zinc regulation occurs in all species of decapod crustaceans.

Zinc is bound so tightly by the proteins in the blood that even in waters containing only a few µg/l of zinc, the concentration gradient for unbound zinc points toward penetration from the water to the blood through the gills. The amount of zinc absorbed in this way increases as the external concentration is increased, indicating that the proteins in the blood to which zinc binds are not normally saturated.[189] So far, no unequivocal evidence has been obtained to show whether or not absorption via the gills is an active or controlled process.

Decapod crustaceans probably receive more zinc than they require, and thus processes for removing extra zinc probably are more important than measures for its absorption. The removal of zinc from the body can occur through the feces, urine, or across the body surface. Losses in the feces depend on whether the animal is feeding and how much zinc is in the stomach fluid. In Homarus, for example, the zinc concentration in stomach fluid is

usually less than 1 μg/g and losses in the feces are very small. In freshwater crayfish, almost all losses of zinc occur in the feces.[192]

The ability to regulate the concentration of zinc--and perhaps the concentration of copper--in the body may give decapod crustaceans a degree of protection in regions where metal pollution is found. The concentrations of zinc and copper in muscle tissue are unlikely to vary, but the hepatopancreas of Cancer pagurus (which is consumed by humans) would probably yield higher concentrations of metal in polluted regions.[186]

Polychaetes

In a study of manganese, iron, and zinc in the Newport River estuary in North Carolina, Cross et al.[339] found that three polychaete worms (Glycera americana, Diopatra cuprea and Amphitrite ornata) showed similar concentrations of metals regardless of large differences in metal concentrations of the sediments in which the worms were burrowing and feeding. Therefore they suggested that these species may regulate body concentrations of trace metals. Bryan and Hummerstone[195] examined Nereis diversicolor, a species closely related to Glycera, and found that whereas the concentration of copper was roughly proportional to that of the surrounding sediment, zinc appeared to be regulated independently of the zinc level in the sediment. It subsequently was found[194] that zinc in Nereis varied by a factor of only 2.7 although they were living in sediments where zinc concentrations varied by a factor of about 30. In these same samples, concentrations of cadmium in the worms were roughly proportional to those in the sediments, indicating that cadmium--like copper--was probably not regulated, whereas zinc was. In experiments to induce toxicosis, worms from sediments high in

zinc were more resistant to zinc than were normal worms. This adaptation of Nereis was ascribed to a reduced permeability to zinc and more effective excretion.[194]

Fish

Theoretical considerations of limited experimental evidence show that fish must obtain zinc from their dietary intakes rather than through exchange with dissolved zinc in the aqueous medium.[726,1222] This conclusion was deduced from the observation that turnover rates of larger body pools of zinc cannot be sustained by inflow rates of zinc from water alone. Such a conclusion again points to the shortcomings of turnover rates estimated from short periods of exposure to radioactive zinc. Estimates of zinc turnover in fish are listed in Table 6-6, along with certain aspects of the experimental approach. Longer turnover times are obtained naturally when the fish are exposed for long periods to zinc-65 in the natural environment.[1332a,1679,1680]

Similarly, the use of short-term exposures of fish to experimental dissolved zinc concentrations in tests for acute toxicosis is of questionable significance. This practice usually provides estimates of lethal levels that can cause death through tissue hypoxia from direct gill damage or precipitation of gill secretions. Under these experimental circumstances, the organisms die before their body pools have equilibrated with the environmental level of zinc, and the significant lethal or sublethal physiologic effects of longer term exposure tend not to be studied.

Zinc concentrations in whole fish are a function of age in certain species, at least in juvenile stages.[337] In croakers (Micropogon undulatus), bay anchovies (Anchoa mitchilli) and menhadens (Brevoortia tyrannus), zinc

concentrations decreased as an exponential function of weight over the range of 0.01-5 g dry weight for the whole fish. In adult bathyal-demersal morids (Antimora rostrata), zinc concentration in white muscle also decreased slightly with size between 200-1,400 g total live weight, but zinc concentration was independent of fish size in muscle of bluefish (Pomatomus saltatrix) between 400-4,500 g.[341] In freshwater carp, the initial zinc concentration in roe decreased with the age of the female, although after gastrulation the zinc concentrations in the larval fish were similar regardless of the parent's age.[1389]

Tissue distribution of zinc-65 in yearling trout was observed after oral administration of a single dose of radioisotope contained in a force-fed gelatin capsule.[1104] Initial high blood concentrations of zinc-65 declined exponentially after 1 day, whereas the isotope was retained for 2-3 days in kidney, spleen, and liver and continued to increase in gills through the fifth day. Muscle, bone, and eye tissues appeared to have equilibrated with zinc-65 by the fifth day and were retaining the isotope more effectively than the other tissues, albeit at a lower concentration than gills, kidneys, or spleen. This experiment suggests that zinc elimination from fish may occur both through urine and gills.

The daily flux of manganese, iron, copper, and zinc was estimated for populations of Atlantic menhaden, spot and pinfish for the summer months in the Newport River estuary in North Carolina.[342] Assimilation efficiencies of these metals by fish were highly variable and dependent on the trace metal concentration in inorganic content of ingested materials. Except for zinc in menhaden and pinfish, assimilation efficiencies were less than 10%. Because a significant fraction of the trace metal ingested is not assimilated, defecation

of unassimilated trace metals by these fish may be a major biologic process in cycling trace metals in highly productive coastal plain estuaries.

By comparing analyses of manganese, iron, and zinc in estuarine organisms, sediments and water, Wolfe et al.[1801] estimated the fluxes of these metals through and within the Newport River estuary and a descriptive model was constructed for the annual movement of zinc in that estuary.[1798] Zinc appears to be retained very effectively in coastal plain estuaries: sediment and detritus reservoirs are large, and efficient recycling occurs through the living organisms. This model was subsequently examined[1799] for sensitivity of the major zinc flows around the detritus compartment toward the other conditions, either measured or assumed, used in the development of the model. The model was most sensitive to phytoplankton incorporation of zinc, microbial zinc assimilation within the detritus compartment, and macrofaunal standing crop, production, and assimilation of zinc. The model was insensitive to changes in Spartina productivity, phytoplankton standing crop, zooplankton standing crop, and productivity and estuarine flushing rate. It is still poorly understood how zinc adsorbed on sediments or incorporated in detritus becomes remineralized and available again for bioaccumulation.

Metabolism of Radioactive Zinc

Zinc-65 introduced into the Columbia River by the Hanford nuclear installations was accumulated by various organisms in the adjacent North Pacific Ocean. Studies of zinc-65 specific activities in fish and prey organisms have produced estimates of intake rates of zinc-65 in the predator fish species and hypotheses of mechanisms of zinc-65 transport in the ocean.[1216,1679,1680] As mentioned,

content of zinc-65 decreased markedly from 1965 to 1971 when the reactors were shut down. Specific activities decreased faster in small individuals of the flounder Lyopsetta exilis than in large individuals, indicating more thorough labeling of longer-lived pools of zinc in larger fish. Specific activity also decreased with increasing depth and with increasing body size for both Lyopsetta exilis and the rockfish Sebastolobus.

From extensive time series of zinc-65 specific activities in Lyopsetta exilis and prey organisms of this species, Vanderploeg[1680] used the model

$$\frac{dS}{dt} = \alpha (\underline{F}(t) - \underline{S}) - \underline{S}\,\lambda,$$

where $\underline{S}$ = zinc-65-specific activity of the predator; α = rate of zinc input as fraction of zinc body burden in the fish; $\underline{F}(t)$ = zinc-65-specific activity of the prey; and λ = physical decay constant of zinc-65. He calculated the daily rate of input of zinc as a fraction of the body burden of zinc in the fish. Values of 0.0027 day^{-1} and 0.0026 day^{-1} were obtained for fish of 22 g and 35 g wet weight.

Zinc-65 from the Columbia River was apparently in a form more available to the Lyopsetta exilis food chain than stable zinc in seawater was to it. Vanderploeg[1679] hypothesized that several mechanisms could contribute to this conclusion. For example, some soluble zinc-65 in the river is rapidly accumulated by plankton and other small particles. A portion of the zinc-65 entering the ocean as suspended particulate material is desorbed[446] and might also be sorbed to particles of marine origin. Herbivorous zooplankton feed on the particles, transform them into fecal pellets, and they, as well as settling particulate matter, deliver high specific activity zinc-65 to the

continental shelf. Distribution of zinc-65 specific activity in the pelagic food web and in fauna at different depths and distances downstream from the river mouth was consistent with these hypotheses. Vanderploeg[1679] found that zinc-65-specific activities in suspended particulate were greater than in the soluble form. His observation is consistent with the existence of stable unreactive complexes of zinc in seawater. Bernhard and Zattera[105] observed that phytoplankton could accumulate zinc-65 and stable zinc differentially if they were available in different physicochemical forms. Piro et al.[1249] found that ionic zinc added to seawater equilibrated with the ionic and particulate zinc already present, but that the amount of complexed zinc remained unchanged. Zinc-65, when added in the form of an ethylenediaminetetraacetic acid (EDTA) complex, however, was distributed among all forms in the same proportion as the stable element. Consequently, the presence of complexed stable zinc in the ocean would promote biologic accumulation of zinc-65 in the ionic form from organisms which selectively accumulate ionic or particulate zinc.

Correlation of Zinc Turnover with Respiration

Retention of zinc-65 has been proposed and used with varying degrees of success as an indirect measure of respiratory metabolism in a variety of organisms.[1069,1159,1160] A very high positive correlation was demonstrated between zinc-65 turnover and oxygen consumption in young plaice, when temperature was the only experimental variable.[417] Similar observations have been obtained with mice.[272,1306] Shulman et al.[1486] found no effect of varying food intake on the turnover of zinc-65 in Menidia menidia and

Fundulus heteroclitus; nor was any effect of varying dietary zinc concentration on zinc-65 turnover found in Lagodon rhomboides L.[727] For zinc-65 turnover to be a valid measure of respiration in the natural environment, both feeding rate and temperature must respond similarly and consistently to environmental changes affecting either function. Thus both feeding rate and temperature--which affect respiration--must be correlated to zinc-65 excretion rate, as has been demonstrated for temperature; and respiration must be correlated with environmental zinc concentration, or the turnover of zinc by the experimental organism must be regulated independently of environmental zinc.

In euphausid shrimp, respiration was poorly correlated to zinc-65 accumulation rates as either a function of temperature or individual dry weights.[503] Turnover of zinc in euphausids was not metabolically controlled and was likely a surface phenomenon, suggesting a strong interdependence on both temperature and zinc concentration. Thus, the utility of zinc-65 loss as a measure of respiration under natural conditions may be restricted to vertebrates. For fish, such a remote technique is especially desirable in natural conditions to avoid the excessive respiration produced by the stress of being confined in experimental respirometers.

TOXICITY OF ZINC TO AQUATIC ORGANISMS

How zinc and other heavy metals are toxic to fish has been studied since the mid-1920's [394, 1490] and the subject recently has been reviewed.[921] Acute heavy metal toxicosis in fish has been attributed to the coagulation or precipitation of mucus on the gills and/or to cytologic damage to the gills. The physiologic mechanism of death from either cause is related to a breakdown

in gas exchange at the gills. Burton et al.[208] studied acute zinc toxicosis in rainbow trout (Salmo gairdnerii) and was able to support the hypothesis that modification of the gas exchange process at the gills creates hypoxia at the tissue level. Tissue hypoxia appeared to be the major physiologic change preceding death once the gas exchange process at the gills was no longer sufficient to supply the oxygen requirements of the fish.

Daphnia magna has been used as a representative of common and abundant zooplankton to study acute and chronic toxicosis produced by several metals.[112] In 48-h tests for acute toxicosis, the median lethal concentration (LC_{50}) for zinc was 100 µg/l when no food was provided and 280 µg/l when food was added to the water. In 3-wk studies for chronic toxicosis, reproductive impairment was found to be a more sensitive measure of toxicosis than survival. The 16% reproductive-impairment concentration was used as the criterion, as values below this level could not be detected from the controls because of variability within groups. The 16% reproductive-impairment concentration for zinc was 70 µg/l. Because the organic material added as food was found to alter the potential for toxicosis, the results may not be directly applicable for any particular water.

Waller et al.[1720] evaluated data from two studies on fathead minnows (Pimephales promelas) populations from three lakes. They concluded that the maximum concentration of zinc to which fish could be continuously exposed should not exceed 1/100 of the 96-h median tolerance limit (TL_{50})--a concentration that caused a 50% reduction in the mean number of eggs laid per female fathead minnow in a laboratory study.

The Environmental Protection Agency has suggested that bluegills be used as a warm water species in bioassays.[1529] McDonald and Heimstra[1009] showed that

bluegills (Lepomis machrochirus) are very agressive and set up dominance-submission relations when confined in aquaria. They found that dominant fish survived an exposure to 32 mg/l zinc longer than submissive fish. A shelter placed in each compartment was found to reduce the number of agressive encounters between fish and reduced the response difference. The results indicate that dominance-submission relations may be a variable for the outcomes of bioassays with fish.

Cairns et al.[216] studied the effects of pH, solubility, and temperature on the ability to produce acute zinc toxicosis in the bluegill sunfish (Lepomis machrochirus). Bluegill sunfish were exposed for 96 h to water-soluble zinc sulfate and water-insoluble zinc phosphate at two temperature ranges (21-24 C and 7-9 C) and two pH ranges (5.7-7.0 and 7.3-8.8). Control fish were maintained in water containing no zinc. No bluegills died in water containing particulate zinc in amounts comparable to the amounts of soluble zinc (13.5, 18.0, 24.0, and 32.0 mg zinc, which produced mortalities of 90-100%. Bluegill mortalities in concentrations of soluble zinc ranging from 10-32 µg zinc/l were zero to 10% at the high pH, whereas at the low pH, where ionic zinc would more readily dissociate from the zinc sulfate, mortality was 100%. Bluegills acclimated to the low temperatures died at a much slower rate, and the time-to-death of the first fish was considerably delayed in comparison to bluegills at the warmer temperature. At the concentration of zinc tested (32 µg zinc/l), the acclimation temperature had no effect on the percentage of fish surviving at the end of the 96-h test period. The acclimation temperature did, however, affect the percent survival of the bluegills at 24 and 48 h.

The toxicity of zinc to the larvae of crab (Carcinus maenas) during periods up to 64 h has been examined.[309] At a concentration of 1 ppm zinc, the median effective time for 50% mortality (ET_{50}) was 47 h. For higher concentrations of zinc, the log of the concentration and the ET_{50} were related linearly. The level of 1 ppm of zinc is about 100 times the concentration found in natural seawater. For periods longer than 47 h, however, the LC_{50} would be cnnsiderably lower and much closer to the level of zinc found in natural seawater. Thus, elevation of zinc levels in confined water to values substantially above natural levels could have a severely deleterious effect on survival of larvae. The toxicity of several metals, including zinc, to oyster embryos in synthetic seawater at 25% salinity and 26 C, also has been tested.[218] For 48-h exposures, 0.075 ppm zinc produced no mortality, and 0.5 ppm zinc produced 100% mortality; the estimated LC_{50} (48 h) was 0.31 ppm zinc.

CHAPTER 7

ZINC IN HUMANS

The roles of zinc in the normal physiology and biochemistry of mammalian systems have been reviewed widely.[362c,504,609-614,1044, 1274,1275,1285,1286,1640] The purpose of this chapter is to describe major aspects of the role of zinc in humans.

ZINC CONCENTRATION IN BLOOD, URINE, AND FECES

Zinc is found in every human tissue and tissue fluid, although concentrations vary in different fluids and tissues. Zinc is present in nuclear, mitochondrial, and supernatant fractions of all cells that have been examined by ultracentrifugation. Total body zinc for a hypothetical 70 kg man may be estimated to be 2.3 g, making it the most prevalent trace metal in tissue.* Table 7-1 compares the estimated zinc concentrations of several tissues in a hypothetical 70 kg man. Clearly, the major amount of zinc in the total body resides in muscle and bone (approximately 90%), although the highest concentrations of zinc is found in tissues from the reproductive tract.

Blood

For convenience, mean serum zinc concentration in humans may be considered to be approximately 100 μg/dl (see Table 7-2 for the ranges involved) and is the same in healthy men and women.[610,669] Reports that differences in plasma zinc occur between mean and women have appeared,[935] but they have not been carefully substantiated. Deviations from the mean of 100 μg/dl may be indicative

*Iron, which exists in higher concentration, is found primarily in blood. For a human weighing about 70 kg, the total concentrations of trace metals are estimated as follows: iron, 4.0 g; zinc, 2.3 g; manganese, 0.2 g; copper, 0.1 g; and all others combined, $\lesssim$ 0.1 g.

TABLE 7-1

Comparison of Estimated Zinc Concentrations

in Some Human Tissues

Tissue	Zinc, µg/g wet wt[a]	Zinc, mg/organ	Zinc, % of total body (70 kg man)
Adrenal	6	0.9	--
Aorta	26	2.6	0.1
Bladder	22	4.4	0.2
Blood	1	6.0	0.3
Bone	66	660.0	28.5
Brain	13	18.0	0.8
Gastrointestinal tract	21	25.2	1.1
Heart	27	8.7	0.4
Kidney	48	19.8	0.9
Liver	27	40.5	1.8
Lung	14	16.6	0.7
Muscle	48	1,420.0	62.2
Ovary	12	0.3	--
Prostate	87	1.7	0.7
Skin	6	30.0	1.0
Spleen	19	3.8	0.2
Testes	13	0.8	--
Thyroid	25	0.4	--
WHOLE BODY	33	--	--
TOTAL	--	2,259.7	98.9

[a] Measurements from Forssen,[491] Soman et al.,[1522b] Tipton and Cook[1620] and Tipton et al.[1621]

of disease or environmental conditions, and such changes will be discussed in Chapters 8, 10, and 11. Estimates of zinc in the serum, plasma, and other fluids of normal adults are set forth in Table 7-2.

Measurements of serum zinc are influenced by many factors. Different techniques of measurement may produce varying results. Sample contamination (a major problem, particularly if glass syringes or collection tubes or rubber or cork stoppers are employed), hemolysis, and the addition of various agents to the sample tend to make values higher. Serum zinc is about 16% higher than plasma zinc,[205] the higher percentage reflecting differences in zinc liberated from platelets[204,482] as well as differences in volume, hemolysis and other unidentified factors.[482] Circadian variation also affects blood zinc levels: values in the afternoon are higher than in the early morning.[693,932] Zinc levels in serum for 2-3 h following ingestion of food have been found to be lower than those measured during the fasting state.[204,1157]

Zinc is also an important constituent of red blood cells, representing approximately 10 times the amount of zinc found in serum. Erythroblasts measured by histochemical techniques do not appear to contain zinc, suggesting that carbonic anhydrase may be found only in erythrocytes.[1588] Systematic chemical analyses have not yet been performed. Zinc also is present in young and mature reticulum cells.[1588] Leukocytes contain more zinc than erythrocytes.[377] Granulocytes, particularly eosinophils and basophils, are relatively rich in zinc (14.2 μg zinc/10^9 cells). Zinc has been incorporated into metamyelocytes, and the content of the metal increases as granulocytes mature.[1588] The content of zinc in peripheral blood granulocytes was found to be about 30% higher than in bone marrow granulocytes.[1588] Most zinc in leukocytes is protein-bound and can be isolated in a purified form.[1660] The function of zinc in leukocytes has not been systematically investigated;[1660] however, leukocytes contain the zinc-dependent enzymes alkaline phosphatase and peptidase, indicating that some zinc in leukocytes may be associated with the activities of these enzymes.[1588,1660]

TABLE 7-2

Zinc Levels in Blood, Blood Fractions, and Urine of Normal Adults

Authors	Number of persons	Mean value ± SD	Range
		Serum or plasma, μg zinc/dl	
Prasad et al.[1277]	19	102 ± 13	80-99
Butt et al.[213]	37 men	157	
	45 women	159	
Gofman et al.[542]	39	98 ± 5	
Parr and Taylor[1210]	6	85 ± 7	
Kahn et al.[809]	97	84 ± 30	
Prasad et al.[1283]	14	104 ± 14	
Helwig et al.[661]	64	91 ± 17	
Parker et al.[1208]	23	90 ± 10	
Davies[360]	30 men	95 ± 13	76-125
	30 women	96 ± 10	
Hackley et al.[595a]	96 men	96 ± 13	72-120
	97 women		86-102
Mahanand and Houck[970]	7	94 ± 29	
Withers et al.[1794]	25	110 ± 21	
Woodbury et al.[1804]	11	85 ± 15	
Meret and Henkin[1031]	82	92 ± 18	
	45 women	90 ± 20	63-147
	37 men	94 ± 18	

TABLE 7-2 continued

Authors	Number of persons	Mean value ± SD	Range
	Whole blood, µg zinc/100 ml		
Dennes et al.[377]	28	560 ± 20	
Brune et al.[181]	7	680 ± 8	
Auerbach[58]	30	734 ± 186	408-1170
Kleinman et al.[852]	15	545 ± 18	
	Red blood cells		
Talbot and Ross[1592]	--	11.8 ± 1.8µg/g blood	8-14.9
Dennes et al.[377]	30	0.97 ± 0.03µg zinc/10^9 cells	
Prasad et al.[1277]	15	12.5 ± 1.2µg/ml blood	11-14.8
Valberg et al.[1651]	57	10.6 µg/g blood	7.7-14
Auerbach[58]	30	1.6 ± 3.3µg x 10^9 erythrocytes	9.6-25
Rosner and Gorfien[1369]	23	12 ± 2µg/10^{10} erythrocytes	8.6-16.1
	Leukocytes		
Dennes et al.[377]	30	14 ± 1.9 µg zinc/10-9 cells	
Frischauf et al.[510]	--	5 µg/g dry weight	
	Units		
	Granulocytes 10		
Szmigielski and Litwin[1588]	bone marrow metamyelocytes	94 ± 8.21	
	juvenile cell	112 ± 7	
	polymorphonuclear cell	139 ± 14	
	peripheral blood	178 ± 12	

TABLE 7-2 continued

Authors	Number of persons	Mean value ± SD	Range
	Urine, μg zinc/24 h		
Vallee et al.[1667]	14	457 ± 120	273-660
Prasad et al.[1283]	5	658 ± 206	
Prasad et al.[1283]	5	658 ± 202	
Helwig et al.[661]	62	525 ± 254	145-1,256
Meret and Henkin[1031]	82	353 ± 207	141-179
	(45 women)	347 ± 322	
	(37 men)	360 ± 128	

Despite the numerous sources of zinc in blood, total circulating hematic zinc represents less than 0.5% of the total body zinc. This amount contrasts with circulating copper in the blood, which represents more than 6% of total body copper but is consistent with the relative percentage of circulating hematic manganese.

Concentrations of zinc in blood plasma vary considerably during the first two years of life,[682,886a] as shown in Figure 7-1. At birth, plasma zinc levels approximate those of normal adults, but within the first few days to a week of life, zinc levels decrease to less than one half of the means shown by normal adults.[682] Zinc concentrations in infant plasma appear to remain at lower than adult levels for the first five or six months of life; then values rise again to the normal adult range, albeit at its lower end. An abrupt fall in serum zinc levels occurring at about one year of age was observed in one group of normal infants.[682] Before 15 months of age, human plasma zinc values remain normal except when influenced by physiologic or pathophysiologic processes. Such changes may be characteristic only of some infants; although clearcut patterns of change have been observed in American infants, few fluctuations have been measured in Western European babies.

In adult life, the circadian variation found in plasma or serum zinc is significant.[204,660a,693,932,1715a] It is generally agreed that zinc levels in serum are at a minimum at 6 a.m., with the maximum occurring at 10 p.m. After 10 p.m., values drop rapidly and consistently. They rise rapidly after the nadir at 6 a.m., and remain relatively stable over the next 8 h.[932] The pattern of circadian variation is somewhat similar to the circadian pattern observed in the secretion of adrenal corticosteroids, although the specific timing of the nadir and zenith is displaced by approximately 4-5 h.

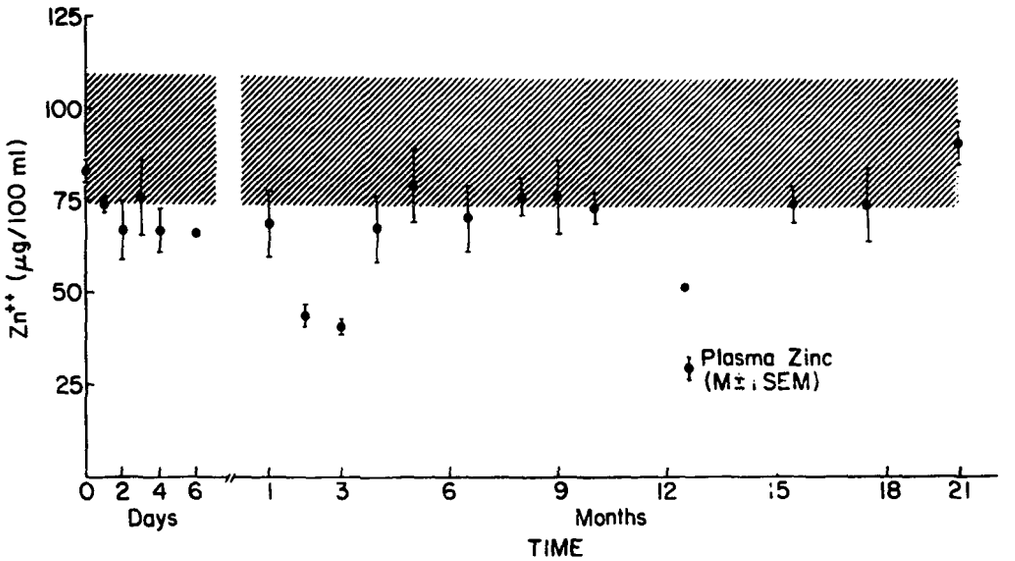


FIGURE 7-1 Total plasma zinc concentrations during the first two years of life. Hatched area indicates normal adult levels (mean $\pm$ 1 S.D.). During the first week of life and at 2, 3, and 12 months of age, plasma zinc concentration was significantly below the lower limit of the normal adult range. All points represent determinations from three or more infants. Where not indicated, all the SEM's were too small to be visible on the graph. Reproduced from Henkin et al.[682]

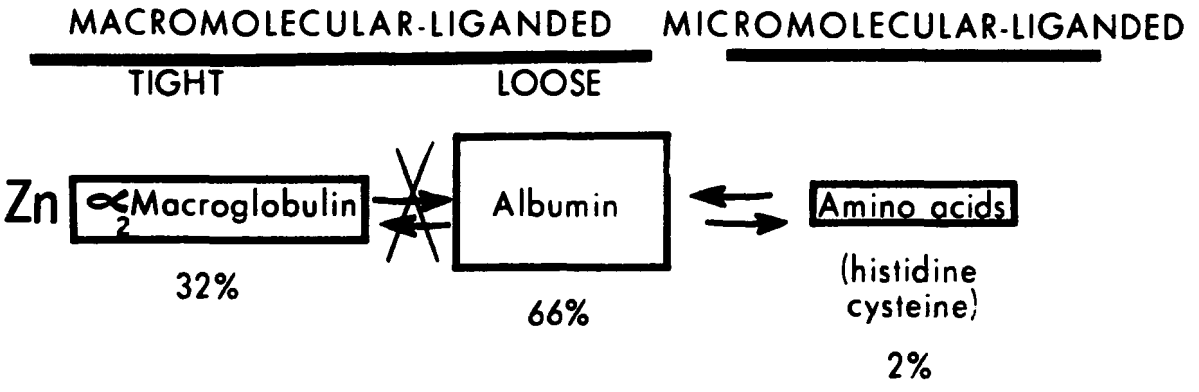


FIGURE 7-2 Bindings of serum zinc. Reproduced from Henkin.[668]

Zinc in serum is always bound to some ligand, as illustrated in figure 7-2. Approximately 43% of the zinc circulating in blood serum is bound to albumin,[537,1205,1692,1802] presumably to one of the histidine moieties of this molecule.[586] However, it has been suggested that zinc binds to the free carboxyl and not to the imidazoles of the histidine residues of albumin.[1226] It is chemically possible for each of the 16 histidine moieties of albumin to bind one zinc molecule,[586] but albumin in human serum is normally undersaturated in zinc.[537] The zinc-albumin complex has an association constant of approximately 10^6 and has been called the major macromolecular zinc ligand.[537,668] This albumin-bound zinc is in equilibrium with amino acid zinc complexes that comprise about 1-2% of the circulating zinc.[537,1278a] This latter group of ligands, called micromolecular zinc ligands, are almost exclusively some form of the amino acids histidine and cysteine.[537,606] These amino acid-zinc ligands are available for transport to all tissues, including body organs, red blood cells, and brain. Histidine and cysteine easily cross the blood-brain barrier.[1166] Porphyrin-bound zinc may serve as another micromolecular zinc ligand,[537,668] although it may not be of particular quantitative importance in normal humans. Small concentrations of zinc-porphyrin complexes have been measured in normal subjects and larger concentrations are present in patients with various types of porphyria.

Approximately one-third of the zinc carried in blood serum is bound to an α_2-macroglobulin.[586,668,1205,1226] This macromolecular zinc ligand, a zinc-protein complex with an association constant greater than 10^{10}, is not in equilibrium with the albumin-zinc complex or the amino acid zinc complexes.[537,668] Zinc is incorporated into this α_2-macroglobulin only in the liver and the complex is metabolized only there.

Zinc in serum may be divided functionally into diffusible or nondiffusible fractions. Diffusible serum zinc is composed of the amino acid-bound zinc and

the freely exchangeable or more readily removable zinc from albumin.[682] This zinc has been quantitatively determined by ultrafiltration.[682] The nondiffusible zinc has been distinguished by ultrafiltration to represent ligands with a molecular weight greater than 50,000. These distinctions have clinical value in the analysis of abnormal zinc metabolism, as noted in Chapter 10.

The distribution of zinc in human serum has also been studied by measuring the partitioning of zinc between the two major macromolecular zinc ligands in serum.[537] As mentioned, albumin-bound zinc is both the major and the more dynamic of the two. However, it is possible to measure the α_2-macroglobulin component of human serum,[1420a] thereby obtaining an estimate of its relative importance. Whereas concentrations of albumin-bound zinc and total serum zinc were highly correlated ($p<.01$) as were the concentrations of albumin and albumin-bound zinc, α_2-macroglobulin-bound zinc was not significantly correlated with either total serum zinc or with the serum concentration of α_2-macroglobulin.

Urine

Approximately 500 μg/24 h appears in the urine of healthy subjects.[669] This zinc content reflects changes in nutrition, physiology, disease, and the environment. In normal subjects, urinary zinc is composed of zinc primarily bound to amino acids and secondarily to porphyrins. Zinc in urine apparently is not correlated with any of the known variables that determine the behavior of serum zinc. Whereas there is a clear circadian variation in serum zinc, none has been observed in urinary zinc excretion.[56] This inability to pinpoint a circadian variation in urinary zinc contrasts with the discovery of a circadian variation in serum and urine copper[932] and may indicate some basic differences in the manner by which the metals are handled by

the kidney. The unhindered passage of zinc-amino acid complexes in normal blood across the renal glomerulus would result in a calculated filtered load of 2 mg of zinc in a normal 24-h glomerular filtrate of 183 l. Thus, the major part of the normal filtered load of amino acid-complexed zinc (and presumably, porphyrin-complexed zinc) must be reabsorbed by the kidney, although the exact nature and characteristics of the reabsorption have not been well studied.

Feces

Zinc in feces represents the major source of zinc lost from the body. The zinc content of feces varies with the zinc content of the diet, and, as such, varies from 5-10 mg daily, and roughly 70-80% of ingested zinc is found in the stool. Zinc in feces also is composed of zinc secreted from the gastrointestinal tract and the bile. Although reabsorption has not been well studied in man, estimates from the rat suggest that zinc is secreted into the gut lumen and that one-third of this zinc normally is reabsorbed.[362b] Zinc in meconium of normal babies varies from 39-569 µg/g dry weight, with a mean of 230 µg/g dry weight. The amount of zinc in meconium was found to be approximately three times the amount of iron or copper and 10 times the amount of manganese.[867c] Infants with cystic fibrosis of the pancreas, meconium ileus, and intestinal obstruction were reported to exhibit lower mean levels of zinc in meconium than did normals.[867c]

Other Body Fluids and Tissues

Zinc in sweat is about 115 µg/100 ml. Under conditions of extreme heat, zinc losses through this route have been reported to increase to as much as 2-3 mg/day.[309a,720a,1288,1585a,1816b] These and other[1355,1474] data indicate that the zinc content of sweat can be greater than that of serum.

Zinc concentration in human milk was found to be higher than that of any other trace element.[1101,1640] The level of zinc in human milk varies with time after delivery.[675] In colostrum the zinc level was found to be 3-5 times the level in milk found 1 wk after parturition,[1640] after which the level fell slowly over the next 6 months.[675,996] It is evident that zinc levels in human milk are carefully controlled, and maternal levels of prolactin have been suggested as an important factor in such control.[675] An average zinc level in human milk 4-6 mo after parturition is 50 μg/100 ml.[675] In milk from cows delivered to market in the U.S., zinc content varied from 1.3-12.4 μg/100 ml with an average of 4.4 ug/100 ml.[1101,1207,1677a] As would be expected, cows' milk stored in galvanized cans was higher in zinc, varying from 55-154 μg/100 ml.[50] Zinc content of cows' milk is reportedly constant all year, but it will vary somewhat with geographic location,[1101] diet, and / other factors affecting bovine metabolism. The importance of the zinc content of milk as an essential nutrient is suggested by a report of zinc deficiency in newborn mice nourished by a mother lactating for a fairly long time after parturition.[412]

Small amounts of zinc also appear in the cerebrospinal fluid (CSF).[669,1660] Zinc has been found in various secretions of the gastrointestinal tract, including saliva,[674,678,684a,1087] gastric juice,[1532,1569] gall bladder bile,[1569] and duodenal and pancreatic secretions.[1569] Zinc is also an important constituent of cerumen. Its concentration is over 1,800 μg/g, roughly 300 μg/g greater than the amount of calcium found, 50 times greater than the concentration of copper,[151] and 5 times greater than the concentration of magnesium.

Zinc has been found in sebum excreted from human skin.[208a,374,1557] By measuring excretion rates of sebum,[344b,1558a] the amount of sebum may be discovered[208a] and its zinc content estimated. In male weanling rats made zinc-deficient, sebaceous gland hypertrophy was observed despite depressed pituitary and testicular function.[1045b] From these observations, hypotheses were advanced that disorders of sebum production such as acne might be related to disorders of zinc metabolism.[374,1557] Patients with acne treated with oral zinc sulfate were said to show decreased sebum production and remission of acne.[374,1557] These findings have not been reproduced.[208a]

In 1940, Eggleton estimated the zinc and copper content of tissues of Chinese subjects by a colorometric technique.[419] Using emission spectroscopy. Tipton and Cook[1620] estimated the concentration of 24 trace elements, including zinc, in normal human tissues from 150 adults who died quickly as a result of an accident (Table 7-3). Many of these values subsequently were confirmed by atomic absorption spectrophotometry.[1451] Although limited sampling made direct comparisons difficult, zinc in tissues of Mideastern and Chinese subjects was higher than in their counterparts in the United States and Africa.[1621]

Forssen carried out similar studies in Finnish and British people who died accidentally or by violence.[491] His results, which are set forth in Table 7-4, are generally similar to Tipton and Cook's, whose findings in American subjects are provided in Table 7-3. No differences in zinc concentrations were observed between men or women and little difference was noted with age, except that zinc levels in the ovary appeared to decrease with age in healthy British women.[623] Zinc in prostate is also known to increase with age, usually in association with prostatic hypertrophy.[1451]

Highest concentrations of zinc were found in human prostate and hair, followed by muscle and liver, and kidney and pancreas; the rest of the metal was distributed

TABLE 7-3

Concentrations of Zinc in Normal Tissue of Adult Americans[a]

Organ or Tissue	Number of Samples	µg Zinc/g Tissue Ash		µg Zinc/g Dry Weight	
		Median	80% Range	Median	80% Range
Adrenal	15	1600	1300-2000	17.6	14.3-22
Aorta	104	1800	1100-2700	68.4	41.8-102.6
Brain	129	760	520-1100	46.36	31.72-67.1
Diaphragm	91	4600	3300-7200	156.4	112.2-244.8
Esophagus	67	2600	1900-4500	98.8	72.2-171
Heart	140	2700	2000-3700	108	80-148
Intestine, duodenum	68	2400	1700-3400	93.6	66.3-132.0
Kidney	145	4500	3200-7400	207	147.2-340.4
Larynx	48	1200	680-2300	111.6	63.24-213.9
Liver	150	3500	2200-5900	129.5	81.4-218.3
Lung	141	1300	880-1800	62.4	42.24-86.4
Muscle	137	4300	2900-6800	180.6	121.3-285.6
Ovary	16	1700	840-2700	74.8	36.96-118.8
Pancreas	138	2300	1500-3500	80.5	52.5-122.5
Prostate	50	7500	3200-19000	337.5	144-855
Spleen	142	1300	1000-1900	65	50-95
Skin	21	780	550-1000	12.26	9.35-17
Stomach	130	2400	1700-3400	76.8	54.4-108.8
Testis	71	1400	900-2200	79.8	51.3-125.4
Thyroid	21	2700	1700-4000	99.9	62.9-148
Urinary bladder	112	3000	1800-4900	69	41.4-127.7
Uterus	32	1900	1300-3400	81.7	55.9-146.2

[a]Data from Tipton and Cook.[1620]

TABLE 7-4

Concentrations of Zinc in Normal Tissue of Adults in Finland[a]

	Ash % of Dry Weight		Number of Samples
	Median	Range	
Brain			
Parietal lobe (grey)	0.093	0.040 - 0.131	20
Parietal lobe (white)	0.052	0.034 - 0.076	20
Hypothalamus	0.066	0.043 - 0.131	16
Cardiovascular System			
Aorta	0.146	0.031 - 0.255	18
Myocardium	0.256	0.151 - 0.435	20
Vena cava	0.174	0.069 - 0.255	14
Respiratory System			
Larynx	0.044	0.015 - 0.123	19
Trachea	0.056	0.023 - 0.103	18
Lung (upper lobe)	0.094	0.036 - 0.188	19
Lung (middle lobe)	0.093	0.017 - 0.216	19
Lung (lower lobe)	0.110	0.012 - 0.152	19
Digestive System			
Esophagus	0.220	0.022 - 0.300	18
Stomach	0.186	0.068 - 0.362	18
Duodenum	0.210	0.124 - 0.346	18
Jejunum	0.220	0.157 - 0.316	18
Ileum	0.220	0.139 - 0.318	17
Cecum	0.189	0.094 - 0.253	18
Sigmoid colon	0.241	0.140 - 0.315	18
Rectum	0.224	0.144 - 0.339	18
Pancreas	0.307	0.133 - 0.780	19
Liver	0.552	0.254 - 1.900	20
Gall bladder	0.097	0.030 - 0.186	12
Urinary System			
Kidney	0.306	0.118 - 0.560	19
Bladder	0.250	0.132 - 0.942	15
Reproductive System			
Testes	0.118	0.082 - 0.159	12
Prostate	0.908	0.303 - 2.870	12
Ovary	0.103	0.066 - 0.139	4
Uterus	0.271	0.066 - 0.298	5
Vagina	0.118	0.072 - 0.159	5
Breast	0.030	0.022 - 0.038	4
Endocrine System			
Adrenal	0.180	0.130 - 0.210	16
Thyroid	0.245	0.062 - 0.513	17
Lymphomyeloid Tissue			
Spleen	0.144	0.068 - 0.211	20
Lymph nodes	0.064	0.015 - 0.123	6
Thymus	0.097	0.081 - 0.108	7
Bone marrow	0.017	0.012 - 0.040	20
Musculoskeletal System			
Skeletal muscle	0.575	0.355 - 0.930	19
Articular cartilage (knee)	0.016	0.005 - 0.051	19
Costal cartilage	0.014	0.011 - 0.040	19
Bone (rib)	0.015	0.011 - 0.028	20
Integument and Fat			
Skin (midventral)	0.073	0.015 - 0.117	13
Fat (under midventral skin)	0.132	0.061 - 0.174	8
Hair	0.784	0.080 - 3.600	6

[a]Adapted from Forssén.[491]

rather uniformly through several tissues.[491] In the endocrine system, the zinc content of the thyroid is greater than that of the adrenal, testes, or ovary.[491] Discrepancies among zinc values in different studies differ primarily in the zinc content of muscle, which varies considerably.

Differences in measurements of zinc content of the same tissues and organs commonly occur, and result in part from the diversity of methods used (emission spectroscopy, atomic absorption spectrophotometry, X-ray fluorescence, etc.) to analyze these tissues. Results have been expressed as μg zinc/g tissue ash, μg zinc/g dry wt, μg zinc/g wet wt, and μg/zinc/g nitrogen. Because soft tissue loses water easily during sample preparation, results calculated on a wet weight basis must be evaluated carefully. Losses and contamination during preparation for analysis can also make it difficult to specify the zinc level in a given tissue with certainty. In addition, the source of the human tissues analyzed is dependent upon such diverse factors as age, sex, physical and clinical well being at the time of demise, drugs used, and etiology and timing of death. For example, tissues analyzed from chronically ill patients had more variable metal levels than were found in patients who died suddenly and unexpectedly.[491,1232] Variability also was reported in tissues according to geographic locations. These data are presented in Table 7-5.

Tissues of rats,[743a,865a] horses,[1451a] and other animals have been analyzed for zinc. Zinc levels in several tissues of ruminants, rats, and humans are compared in Table 7-6.

SPECIFIC ORGAN SYSTEMS

The Reproductive System

Human semen is unique in its high content of about 10-35 mg zinc/100 dl. A positive correlation seems to exist between number of spermatozoa and the

TABLE 7-5

Concentrations of Zinc in Samples of Nine Human Organs Reported from Diverse Geographic Areas[a]

Organ	Area Sampled				
	Finland[c]	U.S.A.[d]	Africa[d]	Near East[d]	Far East[d]
Aorta	0.146[b]	0.180	0.150	0.200	0.250
Brain	0.093	0.074	0.068	0.078	0.092
Kidney	0.306	0.450	0.310	0.330	0.600
Liver	0.552	0.340	0.400	0.380	0.720
Lung					
upper lobe	0.094				
middle lobe	0.093	0.130	0.120	0.160	0.160
lower lobe	0.110				
Myocardium	0.256	0.250	0.200	0.220	0.280
Pancreas	0.307	0.220	0.230	0.190	0.280
Spleen	0.144	0.130	0.130	0.150	0.170
Testes	0.118	0.130	0.190	0.170	0.160

Organ	Finland[c]	India[d]	U.S.A.[e]
Adrenal	0.180	0.290	0.200
Aorta	0.146	0.103	0.180
Brain			
gray matter	0.093	0.102	0.076
white matter	0.052		
Breast	0.030	0.086	--
Esophagus	0.220	--	0.260
Intestine			
duodenum	0.210		0.240
jejunum	0.119		0.220
ileum	0.220	--	0.260
sigmoid colon	0.241		0.260
rectum	0.224		0.330
Kidney	0.306	0.227	0.450
Larynx	0.044	--	0.120
Liver	0.552	0.310	0.350
Lung			
upper lobe	0.094		
middle lobe	0.093	0.120	0.130
lower lobe	0.110		
Muscle (skeletal)	0.575	0.331	0.430
Myocardium	0.256	0.219	0.270
Pancreas	0.307	0.128	0.230
Prostate	0.908	0.107	0.750
Skin	0.073	0.200	0.078
Spleen	0.144	0.126	0.130
Stomach	0.186	0.209	0.240
Testis	0.118	0.100	0.140
Ovary	0.103		0.170
Thymus	0.097	0.094	--
Thyroid	0.245	0.220	0.270
Trachea	0.056	0.103	0.089
Ur. bladder	0.250	0.206	0.300
Uterus	0.271	--	0.190

[a] Adapted from Forssén.[491]
[b] Median values given as % in tissue ash.
[c] Original study by Forssén.[491]
[d] Original study by Soman et al.[1522b]
[e] Original study by Tipton et al.[1621]

TABLE 7-6

Zinc Concentrations in Human and Mammalian Organs[a]

Organ	Zinc, % in ash
Adrenal (sheep)	0.520
Aorta (human)	0.228
Bone (calf)	0.024
Bone (goat)	0.020
Bone (rat)	0.036
Bone (human rib)	0.026-0.037
Heart (calf)	0.187
Heart (goat)	0.195
Heart (sheep)	0.136
Heart (human)	0.117-0.280
Kidney (calf)	0.146
Kidney (goat)	0.170
Kidney (sheep)	0.182
Kidney (human)	0.511-0.517
Liver (calf)	0.300
Liver (cattle)	0.01-0.1
Liver (goat)	0.248
Liver (sheep)	0.346
Liver (human)	0.238-0.690
Liver (human)	0-1.78
Lung (human)	0.23
Muscle (calf)	0.206
Muscle (goat)	0.360
Muscle (sheep)	0.200
Muscle (human)	0.900-1.270
Pancreas (cattle)	0.01-0.03
Pancreas (sheep)	0.225
Spleen (calf)	0.145
Spleen (goat)	0.163
Spleen (sheep)	0.178
Skin (human)	
epidermis	0.580
dermis	0.087
Testicle (calf)	0.013
Testicle (goat)	0.016

[a] Data from Forssen.[491]

level of zinc in semen.[141,1191] Zinc in sperm has been considered an index of male fertility,[423] although this concept is far from proved. Zinc is sperm is removable by incubation with chelators,[739] and it may retard the oxidation of sulfhydryl groups normally present in sperm.[224] The zinc in human semen is approximately 8-10 times as great as the zinc in uterine cervical mucus.[973a]

Zinc concentration may vary within the normal human prostate gland, although it contains a higher zinc content than any other soft tissue in the human body.[109,419,491,937,993] It is of interest that the ventral prostate of rat contains little zinc compared to the posterior portion.[994] These proportions may relate to histologic and physiologic differences observed between these areas. The function of prostatic zinc is unknown, although it may supply zinc to spermatozoa.[1392] On a dry weight basis, prostatic fluid has been reported to be 2-5 times richer in zinc than spermatozoa.[964] The release of zinc has been associated with the activiation of starfish spermatozoa.[514,1075] Cytochemical evidence revealing the influence of zinc in mitosis has been used to explain its presence.[513] Some investigators have tried to correlate zinc in seminal fluid with a zinc-containing protein.[143] Later studies indicated that zinc could be partitioned between high and low molecular weight ligands, although a specific protein could not be identified.[144,145] Studies of boar semen have been repeated in the dog; the zinc in epididymal sperm was loosely bound in both cases.[1393] However, zinc in rat sperm is less available for ethylenediaminetetraacetic acid (EDTA) extraction.[1393] In women, adding zinc to the cervical mucus in the uterus inhibits sperm penetration although copper is a more effective contraceptive agent.

Zinc levels in endometrium are lowest in the late proliferative phase (days 10-12 of the menstrual cycle); they gradually increase thereafter, reaching

their highest levels during the late secretory phase (days 25-27).[596,597] The zinc concentration of cervical mucus of normal menstruating females has also been studied. In menstruating women, zinc in dry cervical mucus varied between 14 and 939 μg/g.[866b] The highest zinc values were found before or just after ovulation (794-939 μg/g dry mucus). A cyclic pattern of zinc concentration was noted in women,[866a,871a] and an observable midcycle peak was measured in the monkey.[1186a] Sharp peaks in zinc in cervical mucus unrelated to menses were thought to come from intercourse, since an average ejaculate contains about 14 mg zinc/100 dl, or about 8-10 times as much zinc as does cervical mucus.[871a]

Kidney

Few data are available on the levels of zinc in kidney other than those reported in studies on tissues in general. In rat kidney, zinc in the cortex was found to be five times higher than in the medulla,[1483] with the highest concentrations occurring in cytoplasm of the cells of the juxtaglomerular apparatus,[1482a] the ciliary edge of the proximal part of the nephron, the ascending part of Henle's loop, and the cytoplasm of the cells in the distal part of the nephron. After unilateral nephrectomy, zinc rises over the first few postoperative days, but after the compensatory hypertrophy occurs the level reverts to normal.[1483] In rats with alloxan diabetes, renal zinc concentration decreased markedly,[1482a] whereas zinc levels were observed to return toward normal after insulin administration. A correlation between zinc content and alkaline phosphatase activity was reported for the renal tubules.[1482a]

Sensory Systems

The concentration of zinc in the human eye is high, although not as high as in the prostate.[964] Human eye tissues may not be as richly endowed with zinc as are some animal eye tissues.[1751,1753,1754] The presence of zinc

in the eye suggests some physiologic role for this metal in the eye or in the visual process.[1752] Indeed, studies with metastable zinc-69 suggest that zinc may be important in uveal and retinal metabolism,[1177] and zinc in the human eye may be related to carbonic anhydrase or retinal reductase activity.[886] The lens is rich in carbonic anhydrase,[66] which may account for the high level of zinc in this tissue.[1602] Rats treated with N-methyl-N-nitrosourea developed retinal atrophy and cataracts associated with increases in zinc content of the eye.[877] A zinc-containing protein has been isolated from the tapetum lucidum of the cat,[332] and the zinc concentration is among the highest found in cat tissues. Zinc has been found in various fluids of the eye, further implicating this metal in some aspect of the visual process.

Zinc has also been found in or near the taste bud[681] and in saliva.[1087] Its absence from the saliva is associated with loss of taste acuity and the appearance of pathologic changes in taste buds.[684] Zinc in saliva has been found in gustin, a metalloprotein that may be involved with providing nutrition to the taste buds.[674] In addition, zinc in saliva has been claimed to reflect zinc nutrition in the body.[678] Because this fluid is easily obtainable, its measurement may be useful in determining some aspects of zinc status in the body.

Brain

Zinc is uniformly present in the human brain.[1424] It is the fourth most prevalent element in the brain, preceded only by sodium, potassium, and magnesium.[333a,344,393,1424] Twice as much zinc is present in the brain as copper. The gray matter of human brain tissue contains about twice the amount of zinc found in the white matter.[491] In 21 brain samples, gray and white matter had 78 and 33.7 μg zinc/g dry weight respectively.[257] Recently, brain regions in experimental animals[627,1143] and humans[641,738] were analyzed for zinc content; zinc was found primarily in the hippocampus, hypothalamus, and mossy fibers

of the cerebellum.[393] Zinc in the cortex was greater than that in the medulla.[1424] Loss of zinc from the brain has been associated with several neurologic symptoms.[679] Zinc has been found in the basal ganglia[1423] as well as in various pituitary fractions.[864] Radioactive zinc accumulates in high concentration in steer pituitary.[456]

Heart

In isolated heart mitochondria zinc alone or with p-chloromercuribenzene sulfonate increased the accumulation of magnesium;[160] it was suggested that zinc altered the permeability of the mitochondrial membrane.[162] Zinc also initiated a vigorous uptake of potassium.[161] Addition of zinc under carefully defined experimental conditions was sufficient to induce the transport of potassium by heart mitochondria.[163,164]

Lung

Little is known about the concentration of zinc in the lung. Few functional studies have been performed concerning changes in zinc levels during physiologic or pathologic changes in pulmonary parenchyma. One report notes that during the acute phase of tuberculosis, zinc levels in lung decrease. (In patients with chronic forms of the disease, zinc levels increase in lung parenchyma.[1809b]) Zinc also accumulates in the damaged pleura of patients with acute tuberculosis.[1809b]

Pancreas

Zinc is found mainly in the islets of Langerhans in the cytoplasm of the α-and β-cells. In other parts of the pancreas, the concentration of zinc is very low. Small doses of alloxan have been shown to increase the zinc content in the pancreas, whereas high doses made zinc disappear from the β-cells; in the α-cells zinc content increased. Thus /zinc may play a role in the function of

cells that elaborate insulin and secrete glucagon.[1482] Although insulin is biologically active when zinc is absent, the metal has been shown to prolong the physiologic action of insulin and adrenocorticotropic hormone (ACTH) preparations, apparently by retarding their rate of absorption.[1274] The relationships between zinc and insulin, and glucagon are thought to be similar.[1274] The role of zinc in ACTH action has not been clearly characterized.

Liver

Very few studies of zinc content in liver of normal humans have been carried out except those in which zinc levels in several tissues were studied. Zinc levels in liver have been studied most often in patients with hepatic cirrhosis, in which condition zinc levels are consistently lower than in normal subjects.[149a,1680a,1680b] From studies of viral hepatitis, it has been found that the production of macromolecular zinc ligands synthesized in the liver is reduced, and the concentration of amino acids found in blood is increased.[683] This shift in equilibrium allows zinc bound to micromolecular ligands to increase, with subsequent hyperzincuria and total body zinc loss.[683,1420a]

Zinc also has been studied in the liver of patients with various types of carcinomas.[863a] Mean normal adult values of liver zinc are 279 µg/g dry weight; zinc levels in patients with cancer ranged from 147-421 µg/g dry weight. The meaning of such a wide range is unclear, although Addink has suggested some relation between high levels of hematic and hepatic zinc.[7] Olson et al. reported low levels of zinc in patients with hepatic tumors.[1171] Zinc in normal liver was calculated to be 38 µg/g wet weight, zinc in the uninvolved portion of carcinomatous liver was 80 µg/g wet weight, and the liver tumors contained 18 µg/g wet weight.

Muscle

The concentration of zinc in muscle represents the largest zinc pool in man. Zinc levels reported in muscle have been quite variable.[491,623,1451,1620,1621] Only a small fraction of the total zinc in muscle is free. The role of zinc in muscle contraction has been investigated but its action is unclear. Zinc binding to glycerol-extracted muscle exerts a strong relaxing effect, probably through interaction with sulfhydryl groups. In small concentrations, zinc produces a marked potentiation of the muscle twitch. At higher concentrations, zinc reversibly blocks transmission at the neuromuscular junction.[1402a,1402b] The amplitude of the contraction of skeletal muscle was reduced in the presence of 0.5 mM zinc and increased with 0.01 mM.[785] The amplitude of cardiac muscle contraction was not enhanced by zinc,[1122] suggesting that the role of zinc is different in skeletal and cardiac muscles.

Hair

Analyses of zinc in hair under some conditions can correlate with the zinc level of serum. Because hair is readily available, it has been commonly used as an index of tissue zinc. However, hair represents "history" as its growth is reflected in its length from the hair shaft. Awareness of this phenomenon is necessary to evaluate any changes in zinc levels along the hair shaft. Zinc fluctuates greatly close to the scalp.[1704] Hair zinc varies in hair of different colors--the highest amounts are in black hair and the lowest in blonde hair--[723,1450] apparently related to the high level of zinc in hair melanosomes and melanoproteins.[723,1549] Zinc may be active in melanogenesis.[476]

Harrison et al. measured the zinc content in hair of 122 normal individuals. The mean dry weight was 178.7 ug/g, the median 171.3 ug/g, and the range was 81-314 µg/g.[642] These values agree reasonably well with other measurements.[411]

It has been claimed that hair zinc levels reflect body zinc levels, but by themselves they are not particularly good indices. Rats fed zinc-deficient diets exhibit lower than normal zinc levels in hair.[1327] Similar low levels of zinc

in hair have been associated with zinc abnormalities in man,[617,1557,1558] but such relationships are more firmly established in animals than in humans.[855] Hair zinc levels are particularly inaccurate in reflecting zinc stores in prepubescent children.[999] Lower than normal levels of hair zinc have also been found in pregnant and lactating women.[620] Hair zinc varies systematically from infancy through the first four years of life[615] and demonstrates changes that generally follow those of serum. At birth, hair zinc levels are similar to those found in adults, but they decrease over the next year of life. Adult levels are not attained again until four years of age. Fluctuations in hair zinc are illustrated in Figure 7-3.

New methods for evaluating the composition of hair take into account the incorporation of metals into the hair matrix during mitosis. Metals also may be adsorbed onto the hair surface or utilized metabolically and incorporated into the structure of the hair shaft. Thus zinc concentration in the cold- and hot-water soluble and insoluble fractions can be determined.[867a]

Skin

Although whole skin does not contain a high concentration of zinc (ranging from 20-1,000 µg/g dry tissue),[545a,1447a,1451,1620] its importance in relationship to the physiologic function of this surface barrier has been emphasized.[307a,307b, 307c,1075a,1232b] Changes in zinc concentration in relation to several dermatologic disease states in man have been investigated by biopsy techniques and neutron activation analysis. Despite the marked variability inherent in tissue sampling, handling, and analysis, lower zinc levels in skin of patients with scleroderma, basal cell carcinoma, and various forms of skin cancer[354d] were found. Application of these findings to clinical dermatology has not been continued

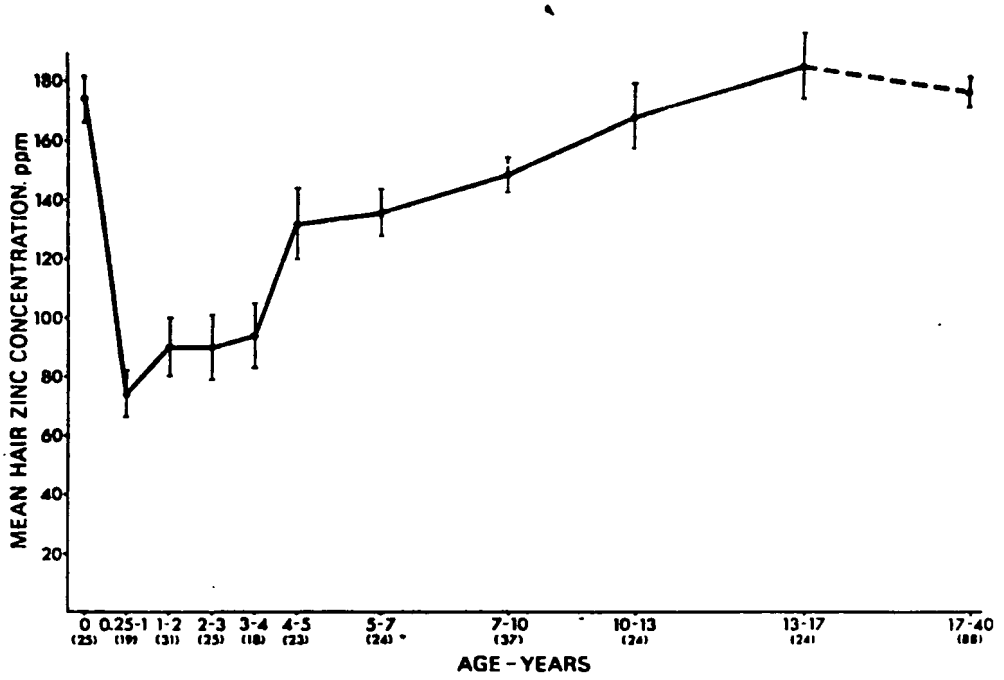


FIGURE 7-3 Mean concentrations of zinc in hair of subjects from infancy to age 40. SEM indicated on graph; number of subjects in each age group is given in parentheses. Reproduced from Hambidge et al.[617]

enthusiastically, although the role of metals, particularly copper and zinc, in collagen formation has been investigated *in vitro*.[729a] The effect of zinc on healing surgical wounds is discussed in Chapter 10.

Bones, Tendon, Nails, and Teeth

Zinc is always found in bones and represents a major body pool of zinc.[17,651,909,1399] Although the function of zinc in bone is not clear, it may aid in calcification as an organic constituent of bone or as an activator of the calcification process itself.[651] Its ability to exchange with bone calcium *in vitro* is well known.[1399,1399a] Even when bone is demineralized, zinc is found in small but constant amounts in the insoluble organic fraction of bone and tendon.[91,1527,433,1527] Thus, zinc could be bound in some manner to the collagen matrix and not merely present in the interstitial fluids.[1527] Indeed, Hsu has postulated a specific role for zinc in collagen metabolism,[729] perhaps in the cross linking of the collagen matrix.

Zinc is present in human rib cartilage.[1437] Concentrations vary with age; it is lowest under 2.5 years of age and after that it rises.[1437] Dwarfs did not show any significant differences from normal subjects.

In teeth, zinc concentration is greatest in the top layer of the enamel, and it decreases in the underlying layers. The deposition of zinc in the tooth enamel probably takes place at the same time fluoride is laid down. Analyses of human enamel showed that zinc was present in all enamel samples but varied widely, ranging from 58-1,550 ppm.[1144]

Pregnancy and Fertility

Changes in zinc metabolism during pregnancy have been studied extensively and reviewed in other portions of this chapter. However, several

investigators have attempted to assess the relationship between zinc metabolism and pregnancy by measuring changes in blood and/or hair zinc during pregnancy. Many changes noted in zinc metabolism during pregnancy are the result of the changing hormonal state, and the shifts in zinc among the various body pools are related primarily to the complex interaction between maternal, placental, and perhaps fetal secretion of several hormones. The dynamics of hormonal interactions have not been fully characterized so that many of the changes in zinc metabolism must still be described as observations determined over time. Changes in blood zinc during pregnancy have been described,[613,616,669,676,797,1164a] as have changes in hair[616] and blood zinc in the fetus and in the amniotic fluid.[669,676] That zinc is retained during pregnancy primarily in the placenta has been hypothesized by Sandstead,[1404] based upon data collected by others.[767,1770a]

A zinc coating has been used either alone or with copper as an effective agent in the control of reproduction following the insertion of a wire, loop, or T-bar device into the cervix in rabbits and rats.[1021,1022] Although the specific role of zinc and/or copper in this process has not been characterized as a chemical event, insertion of zinc and copper wires into the uterus of pregnant and estrous rabbits appeared to increase uterine motility, thereby affecting fertility.[1021] This may be associated with the release of metal ions from the impregnating wire.[1022] Zinc and copper also may interfere with implantation of the blastula.[1601a,1821b] Of further interest is that following the insertion of a copper-T device into the cervix in 16 women there was a much greater increase in endometrial zinc than copper.[1163a]

Changes with Age

Human embryos and fetuses. Zinc has been found in first trimester fetuses, and increases sevenfold between the thirty-first and thirty-fifth day of gestation.[260]

Livers from younger fetuses contain more zinc than do livers from older ones and all fetal values are proportionately higher than adult levels.[1771]

Plasma zinc in human fetuses has been reported to be three times that of adults.[95] Human amniotic fluid also contains zinc; the level may fluctuate during pregnancy.[571a]

Zinc-65 rapidly and easily crosses the placenta and is taken up by fetuses of several species,[455,580,629,630,1605] the ease apparently increasing as gestation age increases.[1605] Some evidence suggests that this increased uptake is dependent upon the fetal liver.[1605] Retrograde transfer of zinc from fetus to mother has also been demonstrated.[1605] Zinc-65 has been found in rat pups even when the only source is the male sire, ultimately from the spermatozoa in which zinc-65 has equilibrated with naturally occurring zinc.[580]

Infancy and childhood. High (>200 µg/g wet tissue) concentrations of zinc have been found in the liver of newborns,[813] which persist until about the fourth month of life. Then levels fall, and the lowest values are reached at age 10; after age 10, hepatic zinc increases gradually until adult values are attained.[813] The zinc content of many brain regions of newborns was lower than that found in adult brains.[641] Zinc contents of infant organs are shown in Table 7-7.

Table 7-8 lists zinc values for some body fluids of children at different ages. Whole blood zinc levels of a newborn child are about 29% the amount found in whole blood of a healthy adult.[852] One reason for the small amount may be discovered in the lower content of zinc and carbonic anhydrase in the red blood cells of the newborn. By the end of the first year of life, the zinc level doubles, and it reaches normal adult values by the second year of life.[957] Zinc levels in blood and hair of infants and children with several physiologic and pathologic conditions have been reviewed recently.[620a]

TABLE 7-7

Concentration of Zinc in Tissues of Infants[a]

Organ or tissue	Age	Sex	Zinc, μg/g wet weight of tissue
Liver	4 h	Female	228
	6 h	Female	105
	13 d	Male	172
	2 yr	Male	80
Kidney	2 yr	Male	33
	6 h	Female	28
	13 d	Male	21
Spleen	6 h	Female	15
	13 d	Male	12
	2 yr	Male	24
Pancreas	6 h	Female	58
Thymus	6 h	Female	16.7
Brain	6 h	Female	7.2
Adrenal gland	6 h	Female	10.7

[a]Data from Parr and Taylor.

1210

TABLE 7-8

Zinc Content of Body Fluids in Children

Authors	Number of Children	Age of Children	Mean value, SD	Range
		Blood serum (S) or plasma (P), μg zinc/100 ml		
Mahanand and Houck[970]	43	1-7 yr	108 ± 15	74-120
Hellwege[660]	13	Abortions	115 ± 22	70-160
	8	Newborn	111 ± 16	78-144
	10	1-12 mo	111 ± 14	83-139
	9	1-2.5 yr	112 ± 15	82-142
	15	2.5-5 yr	111 ± 17	77-145
	22	5-12 yr	114 ± 18	78-150
	16	Over 12 yr	109 ± 11	86-142
Henkin[669]	15	Newborn	83 ± 11	70-100
Henkin et al.[682]	10	21 months	90 ± 16	
		Whole blood, μg zinc/100 ml		
Kleiman et al.[852]	76	Full term newborn	177 ± 6	
	26	Premature infants	147 ± 7	
		Urine, μg zinc/100 ml		
Mahanand and Houck[970]	6	1-7 yr	42	33-55

Changes in organ systems. Changes in zinc with age in several organs have been systematically studied. A simple way to convey this information is through the presentation of figures in which changes in concentrations of zinc in the kidney (Figure 7-4), aorta, brain, and heart (Figure 7-5), liver and lung (Figure 7-6), and pancreas and spleen (Figure 7-7) are charted.

ZINC ABSORPTION

Routes

Zinc is absorbed across several physiologically active membranes, including the gut mucosa, alveolocapillary membrane, and tissue and organ membranes. Absorption across the gut mucosa occurs after oral administration. Absorption across the tissue and organ membranes normally follows gastrointestinal absorption or parenteral administration. Zinc most commonly enters the body through ingestion of food and drink. Inhalation of the metal and its absorption across the alveolocapillary membrane present a specialized type of absorption, because the amount of zinc available via this route is usually small except among zinc workers where metal fume fever or other forms of zinc inhalation may cause clinical problems.* Absorption across tissue and organ membranes is a natural consequence of all forms of presentation. In addition, zinc may be absorbed across the unbroken epithelial membrane of the skin as well as across the broken epithelial membrane after burns or wounds.

Mechanisms

Orally administered zinc is absorbed at several loci in the gastrointestinal tract. Earliest evidence of absorption appears in the stomach as soon as 15 min after ingestion. Although the major site of zinc absorption appears to be

*See Chapter 11.

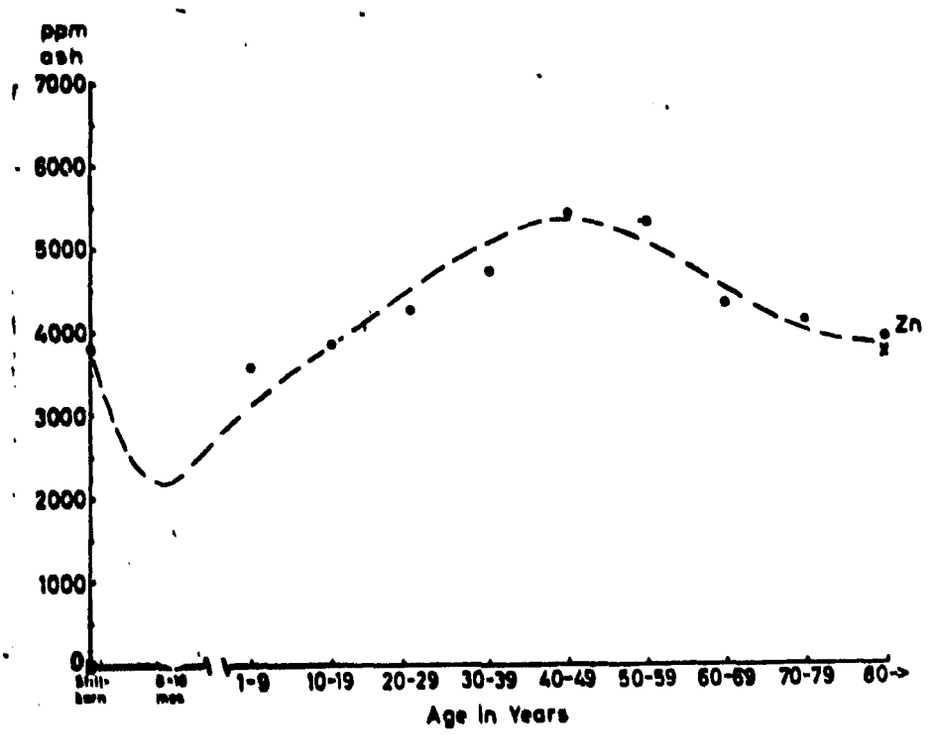


FIGURE 7-4 Mean concentrations of zinc in the kidney in 221 subjects from the United States. Reproduced from Schroeder et al.[145]

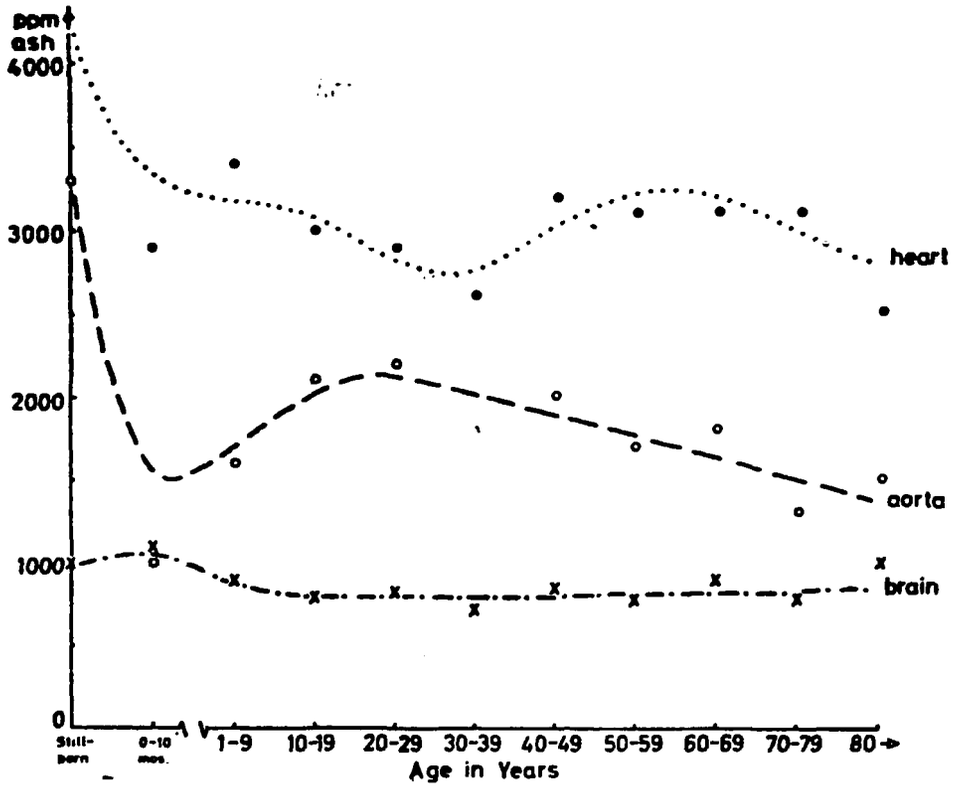


FIGURE 7-5 Mean concentrations of zinc from 121 aortas, 149 brains, and 180 hearts of subjects from the United States. The early decline of zinc in the aorta was the result of measurements from two subjects. Reproduced from Schroeder et al.[1451]

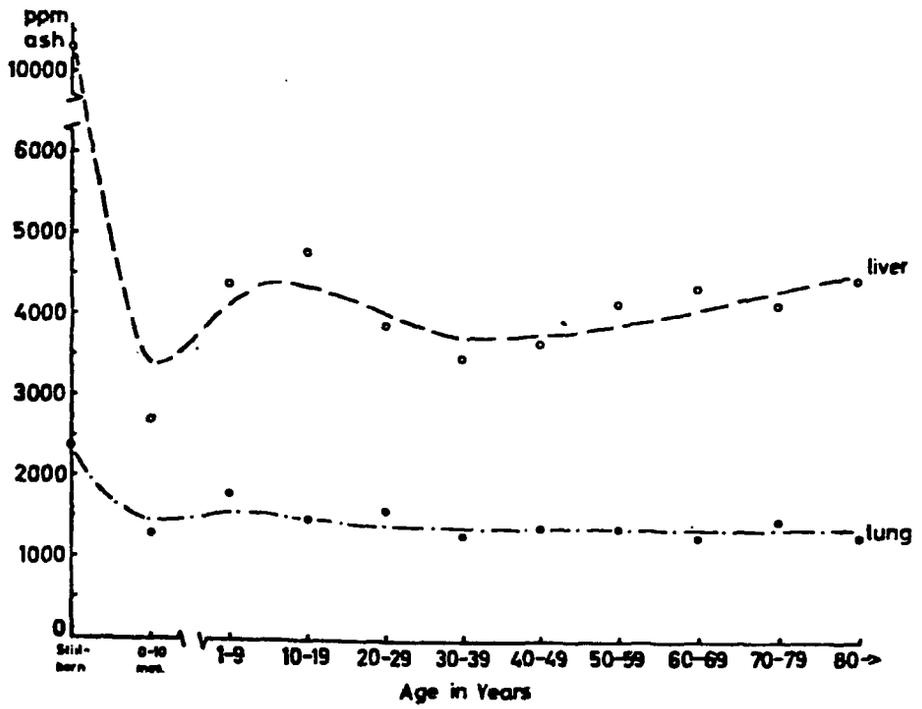


FIGURE 7-6 Mean concentrations of zinc in 231 livers and 188 lungs of subjects from the United States. The low zinc levels in six older infants may reflect the diseases of which they died. Reproduced from Schroeder et al.[1451]

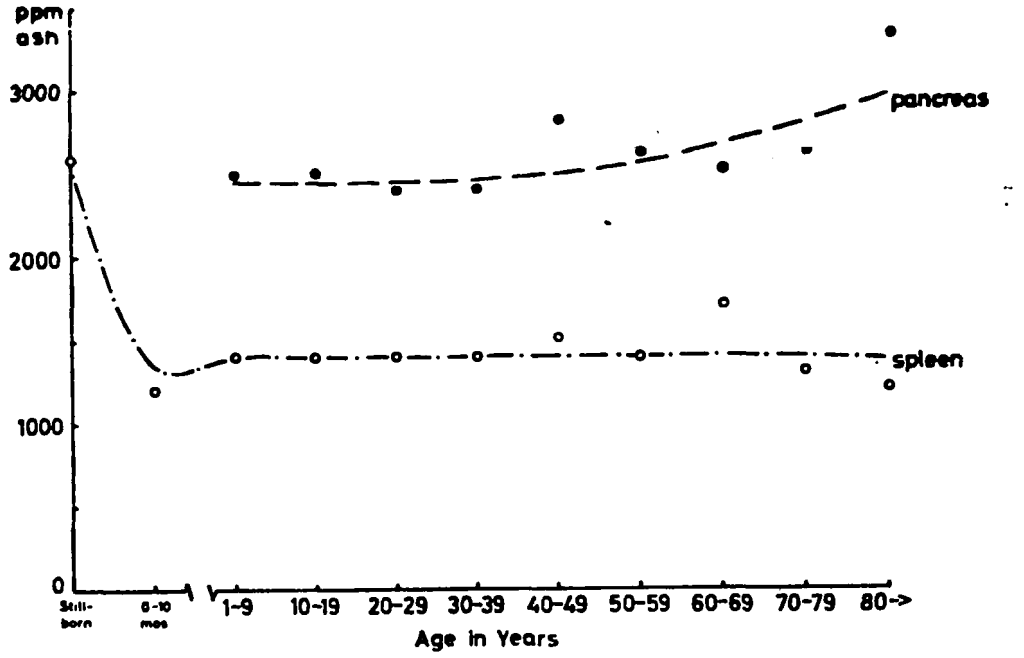


FIGURE 7-7 Zinc in pancreas and spleen in 167 pancreases and 183 spleens of subjects from the United States. Specimens of pancreas from infants were not available. Reproduced from Schroeder *et al.*[1451]

in the second portion of the duodenum, absorption appears to occur at other portions of the small and large intestine.[672a,1035,1036]

The specific manner by which zinc is transported from gut lumen across the gut mucosa or from the mucosa across the gut serosa is not known. However, a consideration of the molecular characteristics of zinc would indicate that the formation of a tetrahedral quadradentate ligand[987] with a small organic molecule is the preferred absorption complex. Our knowledge of this process is limited. The initial event in absorption probably is the formation of a low molecular weight organic zinc chelate; the presence of a zinc-protein complex in the gut has been suggested by several investigators.[1547,1576,1577]

The soluble organozinc complex may be passively absorbed across the plasma membrane of the mucosa of the intestinal villi following first-order kinetics.[1398] Although not established in gut mucosa, this type of kinetics has been suggested in studies of zinc uptake into fish liver slices, where metabolic inhibitors such as dinitrophenol, potassium cyanide, or iodoacetic acid did not influence transport.[1328] Some workers have questioned this passive first-order kinetic process,[1328] but their data--in which an active process is suggested--are not entirely convincing in view of the information supporting the other opinion. The remainder of the ingested zinc forms an insoluble, nondiffusible complex transported and carried in the intestinal products and excreted in the feces. Fecal zinc is composed in large part of all the unabsorbed zinc. However, some absorbed zinc is also found in feces; this amount includes zinc excreted in the bile, in the pancreatic fluids found in the second portion of the duodenum, and fluids composed of back diffusion products derived from gut serosal to mucosal transport. In the rat the serosal to mucosal zinc transport can account for as much as one-third of the total zinc content of the feces. The feces of a man with an average zinc intake of 15 mg daily contains between 10-14 mg zinc.

Many agents affect zinc absorption positively or negatively at the initial stage of absorption. Agents that inhibit formation of the low molecular weight organic zinc complex and thereby decrease absorption include phytate (from bread and other sources),[1157,1328] and soy protein.[367] Other agents that influence absorption (but to a lesser extent due to a smaller oral intake) are peanut, sesame,[906] cottonseed, safflower, and other meals,[1141] other plant proteins,[1517] sphalerites and franklinites,[416] arginine,[716] calcium,[365,489a] particularly when phosphate in high, and phosphates,[691] particularly when calcium is high, other metals,[1669] particularly cadmium,[40] and food itself.[3,822,1421] Oral intake of exogenous zinc even 45 min after a light meal may decrease zinc absorption.*[1421] In the rat, a lowered protein intake was associated with decreased zinc absorption,[1671] but a low protein intake has been associated with increased zinc uptake in man.[1537]

Other factors also influence the manner by which zinc is transported across the intestinal mucosa. They include temperature (as temperature decreases, transport decreases),[1398] concentration of zinc in the external medium (as zinc concentration increases, absorption increases), and efflux of zinc from the tissue itself(e.g., loosely bound zinc will exchange with circulating zinc and become mobilized and metabolized). Concentration of zinc in tissue must be taken into account because it varies inversely to the amount of absorbable zinc. Other factors include the physicochemical characteristics of zinc in relationship to the state of the mucosa,[154] dietary components not previously mentioned (for example, food additives or other substances that act on zinc such as oxidizing or reducing compounds), alcohol,[1564] and the microbial characteristics of the gut. The age of the subject may affect absorption.[688]

*Substances which limit zinc availability by limiting solubility of the zinc itself by complexation with the organozinc complex may be species-dependent.

Adding EDTA and other synthetic chelators to food counteracts the negative effects of phytate and calcium[365,691,741] to some extent,[1508] and other naturally occurring complexing agents produce similar effects.[1157] In states of zinc deficiency, rats and dogs exhibit impaired intestinal absorption of zinc, and it may be considered an anomalous physiologic condition.

Clinical problems of gastrointestinal absorption, including malabsorptive processes of several types, will decrease metal absorption. Zinc can be lost during several disorders, such as excessive renal loss of trace metal because of intrinsic kidney disease or changes in tubular or glomerular function; defective metal binding in plasma of a congenital, acquired, or iatrogenic type; or probably through inborn errors of metabolism, since some aminoacidemias and aminoacidurias are associated with excessive loss of metals.[1495,1496]

A few agents appear to increase zinc absorption in animals by providing or aiding in the fomation of the low molecular weight organic zinc complex; these include histidine, cysteine, and methionine;[537,904,1141] EDTA;[875] and several zinc-sugar complexes.[96] Vitamin C also has been reported to increase zinc absorption with the intake of food.[882] Rats raised in a germ-free environment appear to utilize zinc better than rats raised in an environment with normal bacterial flora.[1509]

When zinc-65 is administered orally, plasma levels peak 2-4 h later;[1345c, 1368,1532,1534,1535,1537] about 50% of the total dose was found in the feces within 15 days after oral administration. The average net absorption of zinc-65 varied between 30-70%, but Spencer observed approximately 50% absorption of the isotope.[1532] She also noted that low zinc-65 levels in plasma reflected low absorption, whereas higher plasma levels reflected greater absorption.[1532] Although high calcium intake decreased zinc absorption in rat, sheep, and other animals, this did not appear to be the case in humans.[1538] Dietary protein does influence zinc absorption in humans. Subjects on a low protein intake had higher zinc-65 plasma levels and lower fecal zinc-65 levels than individuals on a normal

protein intake.[1534,1535,1537] Human subjects on a high protein intake exhibited the expected converse results.

After intravenous administration of a single dose of zinc-65, human liver has been shown to accumulate and retain zinc,[1137] perhaps because of metallothionein in liver.[1538] More than 2 mo after the dose, the liver contained about one-fourth the level found the first day after administration. Zinc is also found in the kidney, spleen, and intestinal mucosa, and zinc levels in lung are similar to those found in the gut.[1532] Pancreas, adrenals, and thyroid show relatively high uptake of zinc-65, whereas prostate, the organ with the highest zinc concentration, is rather low in activity.[1532] In muscle, the turnover of zinc-65 is quite slow; the highest activity is found in cardiac muscle. Bone exhibits low uptake but prolonged retention. Bile concentration of zinc-65 also is low.[1532]

Differences in human blood, fecal, and tissue levels are a function of the route of administration of zinc-65. Following oral zinc-65, the isotope may be found in blood within 15-20 min with peak levels found within 2-4 h. After oral administration, plasma or serum zinc-65 levels are higher than in whole blood. Levels in red blood cells increased for the first 5 days after intravenous injection of a single dose of zinc-65 and decreased thereafter; after 28 days zinc-65 in red blood cells was 2-3 times higher than that in plasma. No matter what the route of administration of the isotope was, most of it was found in the feces, although the amount differed according to route of administration. Following oral administration of a single dose, 66% of the isotope was found in the feces within the first 3 days. After 21 days, 70% of the isotope was found in the feces, 2% in the urine, and 28% remained in the body. Most of the gastrointestinal absorption occurred within 4 h of oral administration. These studies were carried out in older adult males. Thus, little or no information about distribution in young adults or male-female differences is available.

Zinc balance has been studied in only a few small groups of children. In 36 7-10-yr-old girls in 3 different areas in the United States, daily

zinc intake was found to be from 4.6-9.3 mg, with a mean of 6 mg.[437] Protein sources in food were of animal origin in 74% of the subjects. The average fecal excretion was 3.9 mg/24 h, and urinary excretion was 274 ± 69 µg/24 h. Values for fecal zinc correlated with zinc intake. In 2-yr-old children given 4.34 mg zinc, 53% of the metal was retained.[1707] In 8-9-yr-old children given 10.9 mg zinc, 3.8% of the metal was retained.[1707]

Studies of oral absorption of zinc-65 in human subjects provide values ranging from 25-90%. Absorption values found by Richmond et al.[1345c] for four people given zinc-65 chloride orally and calculated by the method of Aamodt et al.[1a,672a] were 34.8, 47.5, 66.9, and 88.8%. Furchner and Richmond[672a] also reported an effect of stable zinc intake on absorption of zinc-65 in one human subject.[515a] Spencer et al.[1534] reported absorption of orally administered zinc-65 in five subjects. They reported values ranging from 27% to 43.9% with an average of 37.5%. The zinc-65 chloride was given as a single, daily dose of 15-25 µCi with breakfast. The values obtained by Aamodt et al. indicate a variation of 19-100%. These values are not inconsistent with the values reported by Richmond et al. and it is probable that binding of zinc-65 to food proteins could account for the lower values reported by Spencer et al.[1534] Comparison of the values from the studies of Spencer[1534] and Richmond et al.[1345c] with those of Aamodt et al.[1a,672a] with taste and smell dysfunction suggest that the patients/they studied fall into three groups--a low absorption group with values between 19-37%, an intermediate absorption group with values between 60-75%, and a high absorption group with values close to 100%. Because only 11 patients were studied by Aamodt et al., the separation of absorption values into three groups was somewhat arbitrary, and the positive results by T-test, comparing the high, low, and middle group, only suggested that these groups were independent.

The long-range component for the 11 patients observed by Aamodt et al. who received oral zinc-65 ranged from 250 to 498 days, except for one patient with a congenital loss of taste, who had very shortened half-times of 143 and 167 days after oral and intravenous administrations, respectively. The combination of low absorption and rapid loss of ingested zinc might be expected to lead to chronic zinc deficiency unless supplemented by a high zinc diet or exogenous zinc.

Two patients studied following oral administration of zinc-65 were found to have an unusual retention pattern characterized by a sharp decrease in the second component of biologic retention. One patient's half-time changed from 288 to 145 days and the other from 350 days to 177 days.[1a,672a] The period of shortened half-time corresponded with the period in which each of these patients had been receiving exogenous stable zinc (100 mg/day). These results imply a washout effect of orally administered, stable zinc. Some evidence for this type of effect had been previously reported.[323]

Retention of intravenously administered zinc-65 by normal human subjects follows a two-component exponential pattern. The rapid component consists of 20% of the injected activity and is lost with a biologic half-time of 8 days. The slow component includes the remaining 80% of the activity and is lost with a biologic half-time of 300 days.[1a] Spencer et al.[1534] reported that two patients lost 25% of intravenously injected zinc-65 with an average half-time of 12.5 days and 75% of the injected activity with a half-time of 322 days. Newton and Holmes[1137] observed that following accidental inhalation of zinc-65 by a human subject, 27% of the inhaled activity was retained with a half-time of 18 days and 73% was retained with a half-time of 453 days.* Richmond et al.[1345c] reported that the slow biologic retention component was 418 days for oral administration to four human subjects. Figure 7-8 compares retention of zinc-65 in man and other species.

*In this study there was a greater uptake and retention of zinc-65 by soft tissues, including liver, than by bone.

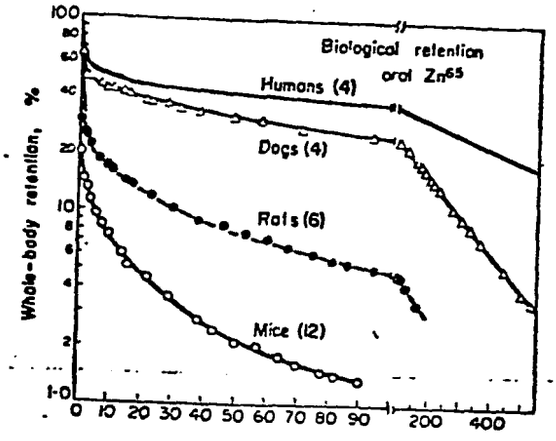


FIGURE 7-8 Retention of orally administered zinc-65 by four mammalian species. Reproduced from Richmond et al.[1345c]

The retention pattern of a patient with cystinuria who took D-penicillamine was different from that previously observed in normal human subjects. This patient's zinc-65 retention after intravenous injection required three exponential components, reflecting a very rapid initial loss: 21.9% of the activity was lost with a biologic half-time of 1.5 days, 17.6% with a half-time of 15.9 days, and 60.5% with a half-time of 235 days.[1a] Increased excretion of zinc-65 during penicillamine treatment has also been reported by Ekberg et al.[422a] and it is probable that the increased removal was a direct effect of D-penicillamine. Moreover, urinary excretion was substantially increased during the first 6 days of the study. The ratio of urinary to fecal excretion was 4.2:1 compared to a ratio of 0.2:1 for 4 patients, 2 with Wilson's disease and 2 heterozygous for Wilson's disease, neither of whom were treated with D-penicillamine.[672a] A urine to feces ratio on the order of 0.25:1 (20% of daily excretion in urine) developed following inhalation of zinc-65.[1137] Spencer et al.[1534] reported variable ratios for urinary to fecal excretion from 0.75:1 to 4:1 in 8 terminal cancer partients, with an average value of 0.49. The variations between urinary to fecal excretion ratios and the limited amount of data available make it difficult to conclude that D-penicillamine increases urinary zinc excretion independent of its effect on zinc absorption.[1534]

After intravenous administration of zinc-65, activity decreased in whole blood and plasma during the first 24 h, followed by increased activity in whole blood and continued loss from plasma. Red blood cell activity increases during the first 5-10 days after injection and then decreases at about the same rate as the plasma thereafter. The biologic half-time is 26 days. Plasma activity initially decreases with a half-time of 3.8 min and more than 95% of the activity is removed by these short half-time components. The data for whole blood of normal subjects were resolved into four exponential components.[1a,672a] Initially, 95% of the injected activity was removed with a half-time of 6.5 min, 2.5% of the activity was lost with a half-time of 80 min, 1.5% of the activity was lost

with a half-time of 2 days, and the remaining 1% was lost with a half-time of about 25 days. These values are consistent with data on whole blood reported for normal subjects and growth-retarded, hypogonadal Egyptian boys,* [1277,1278] and for patients with neoplastic diseases. [1488a] What is striking about the disappearance of zinc-65 from plasma is the extremely rapid loss of nearly all of the activity within the first few minutes after injection.

After intravenous administration of zinc-65, most of the isotope is rapidly taken up in liver. [1a,672a] The rapid uptake phase, completed within the first 20 min, is followed by a slower rate of uptake continuing for 5 h, with maximum activity found between 5 and 24 h. Zinc-65 activity in liver then decreased slowly over the next 100 days. Activity measured over the thigh decreased initially, probably reflecting lost activity in the blood; it then increased during the first 40-50 days after injection as the activity in the liver decreased. The apparent uptake of zinc by the thigh area following release from the liver may imply a biochemical conversion and/or release of the injected zinc to a form stored by muscle or bone.

Although many studies of zinc absorption have been reported, a great deal of uncertainty about the dynamics of zinc absorption still exists. [504, 614,1652] Zinc is still considered to be absorbed poorly and variably. [92]

Data from several zinc-65 absorption studies lead to the following conclusions:

1. Retention (R) of intravenously injected zinc-65 bound to plasma follows a two-exponential component pattern in normal subjects; average retention was:

$$R = 16.2\, e^{-(0.693/6.36)t} + 83.8\, e^{-(0.693/266.5)t}$$

*See Chapter 10.

2. The cystinuria patient treated with D-penicillamine had a markedly different retention pattern, characterized by three exponential elements:

$$R = 21.9\, e^{-(0.693/1.5)t} + 17.6\, e^{-(0.693/15.9)t} + 60.5\, e^{-(0.693/235)t}$$

The major effect of D-penicillamine was rapid removal of 22% of the injected activity with a half-time of 1.5 days. The similarity of the remaining retention pattern to those reported by other investigators and by studies of normal volunteers suggest that the effect of the drug was to remove zinc-65 that otherwise would have been part of the long component of retention.

3. Following injection of zinc-65, activity in whole blood of all subjects [1a, 515a,672a,1345c,1534] decreased rapidly during the first 24 h, then increased to a maximum value between 5 and 10 days after injection, after which it decreased. Activity in plasma quickly decreased to less than 2% one day after injection and then more slowly for the rest of the study period. Red blood cell activity increased for the first 5-10 days after injection, then decreased at a rate similar to that of the plasma thereafter. Although the patient with cystinuria/[given D-penicillamine] always had less activity in cells and plasma than the normal volunteers, no significant differences were observed.

4. Measurements made over the liver area after intravenous zinc administration/[in] patients with taste and smell dysfunction showed a pattern of rapid uptake by the liver, followed by a much slower loss. Maximum liver uptake reached 55-70% of the injected activity between 5 and 24 h after injection. Hepatic retention of injected zinc-65 was described by three exponential components. Normal volunteers showed the following retention pattern:

$$R = 22\,e^{-(0.693/0.60)t} + 48\,e^{-(0.693/7.5)t} + 30\,e^{-(0.693/70.0)t}$$

The cystinuria patient's pattern was very different; the absolute magnitude of the loss from her liver was represented as:

$$R = 59.1\,e^{-(0.693/2.65)t} + 28.0\,e^{-(0.693/15.0)t} + 12.9\,e^{-(0.693/250)t}$$

5. Activity over the thigh initially decreased, probably reflecting loss of activity from the blood. Activity increased after injection for 40-50 days. This pattern suggests that zinc is taken up by bone or muscle once it is released by the liver.
6. The mean/long retention component for patients with taste and smell dysfunction receiving oral zinc-65 was approximately 300 days with a range of 254-498 days.

Zinc Binding in Humans

Zinc complexes of low molecular weight found in serum are thought to be transferred passively across tissue membranes and bound at various concentrations in different tissues, according to the concentration of tissue-binding zinc proteins at specific sites in the tissues. Although metallothionein represents an important binding protein in kidney [807,1304] and liver, [806] other specific tissue-binding proteins may be present. Studies with zinc-65 suggest that liver, pancreas, and kidney retain high levels of this isotope within 96 h of injection, [844,1715] but liver and bone have been shown to contain higher concentrations of the isotope over longer times. Whether these results indicate different binding proteins with different association constants or different concentrations of binding proteins with similar association constants is unknown. This distinction is particularly obvious in the binding of copper to different tissues in patients with trichopoliodystrophy (Menkes's disease) and may be important in normal and pathologic states of zinc metabolism as well.

PHYSIOLOGIC FUNCTIONS OF ZINC

Zinc affects the function of several organ systems. Because zinc is an integral component of DNA polymerase, any rapidly dividing cellular system has an important requirement for zinc. However, several systems requiring zinc metalloenzymes or metalloproteins have a requirement for zinc. In addition, some systems are influenced by the presence of zinc in a manner not readily apparent from their molecular characteristics. This section will attempt to outline the role of zinc in each major system and specify how zinc acts in it.

Zinc directly affects the activities of carbohydrates, lipids, and proteins. Yet in terms of nutritive factors, carbohydrates, lipids, and proteins can affect zinc concentration and thereby change organ function and behavior. Although the role of zinc as a cofactor in enzymes has been well studied,* that zinc is an activator or transducer at cell membranes in intercellular or intracellular systems have only recently been suggested.[285] Indeed, systematic studies of zinc effects were thwarted until reliable techniques were developed in which concentrations of zinc present in the system under study could be easily measured and used to assist in the investigation of metabolic function. The discovery of zinc as a membrane stabilizer, a participant in electron transfer processes, and a major participant in enzyme-substrate interactions, all indicate the important role zinc plays in several physiologic systems.[285]

The Endocrine System

There are many zinc-hormone interactions in which zinc influences hormonal activity at several levels of action, including synthesis, secretion, target-organ binding, and function. Similarly, there are hormone-zinc interactions in which hormones influence the absorption, distribution, and excretion of zinc, its transport in blood, and its action at several levels of organization.

*See Chapter 9.

Zinc-hormone interactions. Zinc has been shown to influence the endocrine system at each of its major levels. Thus, increasing or decreasing the concentration of zinc has been shown to influence hormonal secretion in the hypothalamic-pituitary axis, the anterior pituitary-target gland axis, hormonal synthesis within target glands, and activities at several peripheral organs and target tissues, including hormone binding and utilization.

A surfeit or deficiency of zinc has been shown to influence the endocrine system. Excess zinc will affect the endocrine system, but much less than will lead,[1403a,1816c] mercury,[972b] iron,[575b] cadmium,[518a,1206a] nickel,[888a] and copper.[257a,790a]

It is more common to find that zinc deficiency affects the endocrine system adversely. It is noteworthy that zinc may influence the same or different hormonal systems independently of the mechanism of how the deficiency was produced; that is, from reduced dietary intake or greater body loss.*

Other factors may influence dietary intake of zinc. For example, zinc deficiencies in animals and humans have been reported to be associated with hypogonadism.[382a,1174,1277,1640,1642] This condition has been linked with decreased testicular weight and size, decreased number of spermatozoa, and a late-stage spermatogenic arrest, yet Leydig cells have remained intact. In rats with severe zinc deficiency, growth and development are depressed in the testes,[575b] epididymis, accessory sex organs and pituitary.[423a,488a,1044b] It has been reported that complete testicular atrophy induced in zinc-depleted rats is irreversible,[1044b] whereas in other studies[1045b] gonadotropin administration was associated with gonadal maturation. Zinc supplements have been reported to reverse the signs of the deficiency in all organs, but neither the testes nor the epididymis regain their normal size, function or zinc concentration.[1044b] Similar effects have been

*Zinc-deficient diets and their effects are discussed in Chapter 8.

reported for cattle[1054a] and goats.[1053] However, there was little or no difference in the synthesis of testosterone or dehydroepiandrosterone following the incubation of testicular tissue with hydrogen-3 cholesterol from zinc-deficient or pair-fed control rats.[1272a] These latter findings indicate that the mechanisms involved in this process are not in the testis itself. These results are consistent with those investigators who suggested that the effects of zinc deficiency on the release of pituitary gonadotropins are the major phsiologic factors influencing the changes observed.[1044c] Indeed, lower levels of plasma and pituitary luteotrophic hormone (LH) were found in zinc-deficient rats than in controls although levels of hypothalamic LHRH were similar.[544] Responsiveness to exogenously administered LH and FSH was greater in zinc-deficient than in restricted-fed control rats although a comparison of pituitary or plasma LH or FSH levels between these two groups of rats was not reported.

The influence of zinc on pituitary function has been studied for many years. In 1939, zinc was shown to enhance pituitary activity when a crude extract from the posterior pituitary was injected into frogs and the water-retentive property of the extract was prolonged by adding zinc salts.[150] The action of what might presently be considered to be the pituitary follicle-stimulating hormone (FSH) and luteinizing hormone (LH) was enhanced after zinc salts were added to anterior pituitary extracts and then injected into sexually immature or hypophysectiomized rats.[116,117,997a] More recently, incubating zinc with bovine pituitary extracts has been associated with increases in circulating growth hormone (GH), thyroid-stimulating hormone (TSH), LH, FSH, and ACTH, although the amounts of zinc required to produce these effects were larger than would be necessary for copper or nickel to produce them.[888b]

The specific role of zinc in insulin action has not been clearly defined. When present in the insulin molecule, it is well known that it delays its phsiologic action and prolongs hypoglycemic activity.[1459] Although native insulin does not contain zinc and zinc-free insulin is fully potent, the metal has been useful in the crystallization of the insulin molecule.[1459]

Hormone-zinc interactions. Just as zinc influences the endocrine system, changes in the concentrations of various hormones affect the concentrations of several trace metals in blood, urine, and other tissues. Thus, increasing or decreasing the concentration of trophic pituitary hormones has been associated with fluctuations in body concentrations of zinc as well as changes in the secretion of hormones from target glands. Differences in the hormonal secretion of several target glands also have been associated with changes in zinc concentration. Whether or not releasing hormones in the hypothalamus influence trace metal concentrations is not yet known.

Changing levels of circulating hormones may alter the distribution of zinc in various cellular compartments, promote alterations in zinc-ligand interactions, or alter renal or hepatic handling of trace metals. These changes generally accompany changes in the body concentration of zinc either because they produce a shift to tissue compartments not normally associated with such zinc concentrations or they may increase urinary and fecal loss, thereby bringing about total body loss.

Effects of growth hormone. Increased concentrations of circulating growth hormone, which occur in patients with untreated acromegaly, have been associated

with reduced serum zinc and greater urinary zinc excretion.[667] Treating these patients with surgical hypophysectomy or X-irradiation, procedures which lower circulating growth hormone levels, also decreases urinary zinc excretion, increases serum zinc concentration, and slightly decreases serum copper concentration.[667]

Conversely, decreased concentration of circulating growth hormone, which occurs in patients with untreated, isolated growth hormone deficiencies, has been associated with elevated levels of serum zinc and decreased excretion of urinary zinc.[667] Treating these patients with exogenous growth hormone lowered the elevated serum zinc level and elevated the previously reduced urinary zinc excretion.

To demonstrate these interrelationships further, changes in trace metal metabolism were studied following the intravenous administration of arginine hydrochloride to patients of short stature to evaluate growth hormone release.[667] In one subject with low plasma levels of growth hormone, serum zinc concentration was elevated and excretion of urinary zinc was depressed. Following arginine administration, growth hormone concentration in blood increased, whereas serum zinc levels fell; urinary zinc excretion increased in temporal relationship with the rise of growth hormone in plasma.

These results suggest that growth hormone levels in blood are directly related to urine levels of zinc excretion. These changes may be related to the manner by which growth hormone either directly and/or indirectly affects the binding of zinc to its macromolecular and micromolecular ligands in blood, thereby affecting the urinary excretion of zinc.[537,668] However, the specific relationships between total serum zinc concentration and plasma growth hormone levels are not entirely clear, because there was no significant correlation between these two variables when the data obtained were plotted.[667] The lack of significance may indicate that blood growth hormone levels do not influence macromolecular liganded zinc[668] as

much as zinc bound to albumin or peptides.[668] Therefore, total serum zinc may only reflect some of the physiologic and biochemical changes that occur in hormonal disorders.

Effects of progesterone. Little or no change in serum or urinary zinc occurs after rats receive small amounts of progesterone. However, serum zinc concentrations decrease following administration of large amounts of progesterone to rats.[1415] In women receiving various drugs to control fertility, serum zinc concentrations may be lowered, whereas serum copper concentrations are elevated.[613] The mechanisms by which these changes occur have not been well studied.

Effects of other gonadal hormones. In adult rats with gonads removed, serum zinc concentrations are uniformly low.[1415] They increase with the administration of the appropriate male or female gonadal hormones.[1415] The pattern of change in serum zinc has been well studied in women and rats during their menstrual or estrus cycle,[599, 1203, 1415] pregnancy,[599, 676, 797, 839, 1203, 1548] and during pseudopregnancy in rat.[1415] Each condition demonstrated an inverse relationship between estrogen and progesterone and serum zinc concentration. In these studies little influence of pituitary gonadotropin on zinc metabolism in gonadectomized rats was observed,[415] although the influence of pituitary hormones on male hypogonadism in rats is well known.

Zinc, as noted, is present in the placenta of pregnant women, and levels change during pregnancy. In one study, at term, the zinc content of the placenta was high, representing 1-2% of the total zinc content of the pregnant female.[1548] In another report,[480] the zinc content of the placenta was highest 6-12 wk into the pregnancy and it decreased thereafter. The role of zinc in the placenta is really not known. However, it may be related to its high level of alkaline phosphatase,[707] an enzyme that increases in serum as pregnancy proceeds.[253]

Changes in serum zinc have been studied during the menstrual cycle of normal females with contradictory results. However, there was one indication that serum zinc concentrations did exhibit changes during the menstrual cycle, with the highest level coinciding with the time of ovulation.[599] Whether any specific relationship between ovulation and zinc metabolism exists is still unanswered.

In humans, the influence of gonadal steroids and their analogues has been most clearly demonstrated in the effects of these agents on lowered serum zinc levels.[611,613,1415] No specific contraceptive effect was imputed to the changes in serum zinc levels, but the possible importance of their coexistence should be noted.

The exact role of zinc in testicular function is not known. In rats fed zinc-deficient diets, testicular atrophy eventually developed.[80] In zinc-deficent rats, alterations in protein and nucleic acid metabolism in the testes were found.[962] Studies of the effects of gonadotropins and testosterone on zinc-65 uptake in male rats also have been carried out.[1367] Administration of FSH decreased zinc-65 uptake in tissues of the reproductive tract as well as tissues not involving gonadal function.

The nature of the synthetic defect in testosterone metabolism from zinc deficiency has not been clearly established. It is clearly demonstrated that sperm contain the highest concentration of zinc of any bodily tissue per milligram protein.[1338] High zinc concentrations have been found in the heads of individual spermatozoa by X-ray microprobe techniques,[604a] although this finding has been ascribed by later workers to contamination with seminal vesicle fluid.[511a] Zinc is highly concentrated in ova of all species, but its purpose, except as related in some general way to growth and development, is not clear.

Changes in the metabolism of pregnant women produced changes in zinc content of some tissues.[767a,1404,1770a] In eclampsia of pregnancy, zinc levels decreased to about 10% of their normal amounts. Infectious processes or neoplasia complicating pregnancy raised the content of placental zinc. It has been

suggested that loosely bound zinc may affect the epithelial tissues of the uterus. Some toxic states in pregnancy are associated with the loss of zinc by the placental epithelium.[1138]

Effects of insulin. Because of the high content of zinc in the islets of Langerhans and the prolongation of action of insulin by zinc, a relationship between zinc and diabetes has been sought. In patients with diabetes mellitus without proteinuria, urinary excretion of zinc was reported to be above normal. In patients with diabetes and proteinuria, urinary zinc excretion is understandably higher than in normal subjects.[310,717] Plasma zinc levels of fasting diabetics have been reported to be higher than in normal subjects, and diabetics of more than 10 years' duration have been reported to have plasma zinc levels higher than those in the earlier stages of the disease.[310] Whether or not the extremely high values in diabetics of more than 10 years reflected prolonged therapy with zinc insulin was not determined.

However, in recent studies, no correlation could be obtained between plasma levels of zinc, glucose, or glycosuria.[361] Nevertheless, these investigators demonstrated a striking fall in plasma zinc after loading with oral glucose.[367,717] On the basis of fat-free pancreas tissue, little difference in zinc content of the pancreas from normals or diabetics was measured by some investigators,[422] although others estimated that the zinc content of the pancreas was one-half that of nondiabetics.[1460] These problems have not been resolved yet.

In oral and intravenous glucose tolerance tests, glucose rose as plasma zinc concentration fell;[361,717] however, the curves were similar for both normal subjects and diabetic patients. In zinc-deficient rats, glucose intolerance was observed after an intraperitoneal glucose load but not after an oral glucose load.

Thus, insulin has a biologic action independent of zinc. However, zinc forms a physiologically active compound with insulin that delays its physiologic

action and greatly prolongs the hypoglycemia.[1459] Many studies have attempted to elucidate the structure of zinc insulin. Imidazole groups are implicated in the binding of zinc to insulin.[165,1570] The ratio of zinc to the insulin monomer is approximately 0.5 g atom/mole;[1271] in actuality, there are 3 zinc atoms/insulin hexamer. Measuring the kinetics of photooxidation suggested that the rate of photooxidation of zinc-free insulin was considerably faster than that of zinc insulin.[1746] Analyses by X-ray techniques have offered evidence of the structure of crystallized insulin from different species. For example, bovine insulin crystals are rhombohedral.[391,632] Zinc is useful in the crystallization of insulin: if the ash content of crystalline insulin is lowered it will not crystallize again unless a metal salt is added.[1457a,1459]

Effects of parathyroid hormone. Patients with untreated hyperparathyroidism exhibited increased urinary zinc excretion.[971a,971b] However, the mean concentration of total serum zinc in these patients was normal. Following surgical treatment of the hyperparathyroidism, urinary excretion of zinc decreased among patients whose parathyroid hormone (PTH) levels reverted to normal; in patients with persistent hyperparathyroidism, no decrease in urinary zinc occurred after surgery.[971b] These clinical results suggest that parathyroid hormone was directly related to body zinc loss from urinary excretion. The results were partially confirmed by studies in which purified parathyroid extract was administered intravenously to two patients in whom urinary zinc excretion increased two- to threefold during the second hour after infusion. However, their urinary cyclic adenosine 3':5'-cyclic phosphate (AMP), which increased after the first 15 min, had returned to baseline levels.[971b]

There are several possible ways PTH could have produced these results. For example, increased metal excretion observed in patients with untreated primary

hyperparathyroidism or after PTH infusion could have been the result of an aminoaciduria induced by the PTH excess. About 36% of the patients with primary hyperparathyroidism have been reported to show a generalized aminoaciduria,[346a] and intravenous infusion of PTH increases the urinary excretion of histidine and other amino acids by about 50% within 2 h.[1491a] Since oral administration of histidine has been shown to increase urinary excretion of zinc, these results are consistent with the concept that the blood histidine strips the zinc normally complexed with albumin, forming histidyl-zinc ligands that are readily excreted in the urine.[668,679] Another possible effect relates to the increased bone turnover commonly observed in patients with primary hyperparathyroidism.[800a] Since a large proportion of body stores of these metals is located in bone,[1640] the increased urinary excretion of these metals in hyperparathyroidism may be an index of bone turnover. The increased urinary excretion of copper and zinc observed in acromegaly,[667] a disease in which bone turnover may also be increased, may also reflect this phenomenon.

Effects of adrenocortical steroid. Abnormalities of adrenocortical steroid metabolism in man and animals have been linked to changes in copper and zinc metabolism.[670] Adrenalectomy or adrenocortical insufficiency from several causes, including idiopathic Addison's disease and hypopituitarism, have been accompanied by increased serum zinc concentration, decreased urinary zinc excretion and increased retention of zinc in several tissues.[670,677] Exogenous replacement of adrenocortical steroids in both man and animals corrects these changes by normalizing the serum concentration, urinary excretion and tissue retention of the metal.[479a,670]

Circadian changes in copper[1091b] and zinc[932] levels in blood have been associated temporally with the circadian variation observed in the levels of 17 hydroxycortical steroids in blood and urine,[667] although administration of

exogenous carbohydrate-active steroids did not abolish the circadian variation observed for these metals.[932]

Conversely, elevated endogenous secretion of adrenocortical steroids, which occurs in Cushing's syndrome or in patients with adrenocortical carcinoma, has been associated with decreased plasma zinc, increased urinary zinc excretion, and decreased tissue retention of zinc.[670,677] These changes are a consequence of increased excretion of metals. Any treatment of these diseases that helped the excessive endogenous adrenocortical steroid secretion brought about the return of serum copper and zinc concentration to or toward normal, a reduction in urinary excretion of zinc and an elevation in tissue levels of zinc.[670,677]

Similar changes in patients with hypersecretion of adrenocortical steroids were observed following oral administration of exogenous carbohydrate-active steroids to normal volunteers.[670] After five days of being dosed with 50 g prednisolone daily, the subjects' serum zinc concentration decreased and urinary zinc excretion increased.

These studies indicate an inverse relationship between adrenocortical steroid levels in blood and serum zinc concentration, and a direct relationship between blood levels of adrenocortical steroids and urinary excretion of zinc. These changes may be related directly to some adrenocortical steroids affecting the production of increased ultrafilterable zinc in serum.[669,670] Such changes previously were associated with decreases in serum zinc concentration and increases in urinary zinc excretion.[670] However, increased renal excretion of copper after adrenal corticosteroid administration[1748a] may be linked to changes in glomerular filtration produced by adrenocortical steroids. These fluctuations may also affect serum levels and urinary secretion of zinc and copper. It has recently been demonstrated in mammalian cell cultures that zinc uptake was stimulated by the adrenocortical steroids hydrocortisone and prednisolone.[1205] The action of these

steroids on zinc accumulation was thought to be caused by stabilization of cellular membranes, since zinc protects membranes from fragmentation during subcellular fractionation.[1205]

Prostate and Gonads

Zinc concentration in the prostate, seminal vesicles, and other associated structures is higher than in any other soft tissue of the body. Table 7-9 lists zinc concentrations in the male reproductive tract. In Sanyal and co-workers' studies of rats, removal of zinc caused irreversible degeneration of the seminiferous tubules of the testes. Degenerative changes were also found in the prostate and seminal vesicles after zinc depletion, but they were reversed after administration of androgens, chorionic gonadotropins, or zinc supplements.[1412a]

Zinc concentration in prostate tissue undergoing various types of pathologic changes has been reported to vary widely from normal. Verrilli *et al.* attempted to make a diagnosis of early prostatic carcinoma and a differential diagnosis of prostatic nodules by studying the differential uptake and distribution of zinc-65 throughout prostatic tissue; their efforts were unsuccessful in making a selective differentiation.[1685]

TABLE 7-9

Zinc in the Male Reproductive Tract[a]

	µg/g Fresh Tissue		Number of Organs
	Mean	Range	
Prostate gland			
Lateral lobes	210	124-399	11
Posterior lobe	190	123-360	10
Epididymis	115	73-189	9
Testes	61	41-130	9
Ampulla of vas deferens	153	57-295	12
Seminal vesicles	108	39-305	17
Striated muscle	68	54-86	10

[a] Data from Lindholmer and Glauman.

Zinc in prostatic cancer, studied by the assay of individual tissue sections, confirmed previous findings of lower than normal levels of zinc in this tissue.[1443] However, a high zinc level in prostate has been observed in benign prostatic hyperplasia.[1443] Normal human prostatic fluid contains a high level of zinc, and in cases of benign hyperplasia the most elevated zinc levels were in prostatic fluid, an extracellular fluid, instead of within cells. The *in vitro* uptake of zinc-65 was highest in cases of prostatic cancer and lowest in prostatic hyperplasia. An inverse relationship has been observed between zinc concentrations and the *in vivo*[594] and *in vitro* binding capacity of zinc-65 in different pathologic stages of prostatic tissue.[593] Further evidence showed that zinc within the prostate was bound to sulfonated-acid mucopolysaccharides: autoradiographs indicated that zinc binding areas corresponded to areas of acid mucopolysaccharides.[593]

A zinc-binding protein has been isolated from human benign hypertrophic prostate tissue.[659] Analysis of amino acids in this protein reveals a very high molar ratio of histidine. The importance and specificity of this finding awaits further investigation.

Because of the wide variation of zinc reported in prostate--ranging from 30 to 2,315 μg/g dried tissue--[709,1443] several investigators attempted to correlate zinc concentration in prostate by the histologically estimated ratio of zinc in epithelium and stroma.[835a,948b] However, the results were not very useful. Gonic *et al.*[544a] stated that zinc prostate concentration varied inversely with the amount of prostatic fibromuscular stroma. In an attempt to establish some standard, several investigators attempted to correlate levels of zinc and β-glucuronidase activity in prostatic tissue.[1091a,1091c,1143a] Their results indicated a good correlation between zinc concentrations and β-glucuronidase activity in noncarcinomatous tissue but not in carcinomatous tissue.

Several investigators, using techniques correlating zinc levels with histochemical enzyme activity, confirmed the extremely low levels of zinc in carcinomatous prostatic tissue (40-50 μg/g tissue wet wt), [593,1091c] and reached fairly good agreement about the level of zinc in noncarcinomatous prostatic tissue (135-140 μg/g tissue) [140a,1091c] and in prostatic fluid (480-690 μg/g tissue). [835a, 948b,993,1091c,1766a]

Since patients with benign prostatic hypertrophy exhibit elevated levels of zinc in prostate, treatment with several chelating agents has been suggested as a way of producing decreases in the symptoms associated with this condition and decreases in the size of the prostate. [593,948b,1008a,1443] These results have not been tested in controlled clinical trials, yet they do appear encouraging. These effects could be explained as an effect of the chelating agents on the noncarcinomatous prostate cells such that hyperplastic areas of the prostate are shrunk.

Low levels of zinc in prostate have been observed not only in carcinoma of the prostate, but in patients on zinc-deficient diets. Low levels of zinc also have been observed in the prostatic fluid and testes of patients with zinc deficiency. In recent years the incidence of hypogonadism among zinc-deficient subjects has been noted, [383,1174,1277] and the relatively high concentrations of zinc in the prostate gland and in sperm have been related to the role that zinc plays in gonadal function. However, this / interaction has not been clearly defined. In a double-blind study of zinc-deficient Egyptian dwarfs given either placebo or supplemental zinc, no differences in growth or gonadal function could be demonstrated between the two treatments. [1410] Studies have been carried out in which gonadal weight and zinc content of rats fed zinc-deficient diets have been compared with similar measurements made in controls fed restricted amounts of food. Interpretation of this study is difficult since 3-4 rats were grouped in each cage, [993] thereby

greatly increasing the variability of the results. Exogenous testosterone or pituitary gonadatropins also have been given to rats fed zinc-deficient diets, but as pair-fed controls were not studied simultaneously [1045b] interpretation of these results is limited. However, there is little question that decreased testicular weight in zinc-deficient rats is a consistent feature associated with zinc deficiency.

Differences in morphology between the testes of zinc-deficient and pair-fed control rats indicate a true spermatogenic arrest in the zinc-deficient animals in one study. [1272a] Although destruction of the germinal epithelium was not observed, which others had noted, [1045b] decreased tubular size was apparent, as was the absence or marked diminution in number of sperm. The presence of Leydig cells in the testes of zinc-deficient rats was noted in one study. [1272a] Vacuolated nuclei of the spermatocytes were reported in this same study. [1272a] Zinc deficiency also has been observed to reduce the diameter of testicular tubules and to produce atrophy of the germinal epithelium up to the spermatogonia stage with sloughing of cells into the tubular lumen. [1045b] This latter change became more apparent as the zinc deficiency was prolonged. It must be pointed out that starvation, i.e., pair-feeding *per se*, apparently did not produce significant histopathologic changes in the rat testes. [1272a]

That zinc deficiency alters gonadal function is clear; however, the manner by which it occurs is unclear. Either zinc is involved in some aspect of gonadal function other than testosterone synthesis, e.g., in the synthesis or release of pituitary LH or FSH, or the inanition produced by zinc deficiency impairs gonadotropin release. Although little data are available to support either hypothesis, previous studies suggest that zinc and gonadatropin function may be interrelated. Exogenous administration of gonadotropins to zinc-deficient rats increased the weight of epididymis [1045b] but did not alter testicular zinc concentration.

Although the results of these studies were quite variable and pair-fed rats were not studied, some gonads of zinc-deficient rats appeared to mature once gonadotropins were administered. These animals' pituitaries were small in size and more morphologic "gonadotrophs" were recognized when compared with those of normally fed controls. In some ways the morphologic appearance of the testes of zinc-deficient rats is similar to that of patients with hypogonadotropic hypogonadism; i.e., a spermatogenic arrest accompanied by spermatogonia and Leydig cells is observed in both conditions.

In addition to these effects of zinc deficiency on testicular function, zinc in high concentrations can be spermicidal.[1766]

Renal Function

Extrapolations from model studies suggest that the unhindered passage of zinc-amino acid complexes in normal plasma across the renal glomerulus would result in a calculated filtered load of 2 mg zinc in a normal 24-h glomerular filtrate of 183 l.[537] Observations of several investigators[537,668,1503,1551,1666] indicate that under normal conditions about 0.5 mg zinc is excreted in urine daily. Thus, the major part of the normal filtered load of amino acid-complexed zinc must be reabsorbed by the kidney. Although earlier workers suggested that urinary zinc excretion was not a function of intake, this notion may require further evaluation because many recent studies have questioned it.

If plasma zinc shifts such that more or less zinc is complexed with micromolecular ligands,[668] normal zinc transport and storage processes may be altered. An increase in micromolecular zinc ligands (amino acid-zinc ligands) crossing the renal glomerulus could lead to excessive loss of zinc through urine. Albuminuria would lead to hyperzincuria as well.

The manner by which zinc is reabsorbed by the kidney is not clear, although analyses by laser microprobe and histochemical techniques have suggested that a high concentration of zinc resides in the proximal tubule of the kidney, where reabsorption may be expected to take place.[668,1483] If reabsorption of zinc occurs, then the role of metallothionein as an agent in the conservation of zinc could be an important one. However, agents such as carbonic anhydrase and alkaline phosphatase--both zinc-containing enzymes--have been shown to occur in high concentrations at the brush border of the renal tubule.[541a] Thus, although zinc loss is controlled primarily through size discrimination at the renal glomerulus (which inhibits the loss of macromolecular-liganded zinc moieties), specific properties of other substances and enzymes influence the tubular reabsorption of a very large amount of zinc.

Schroeder has shown that kidneys of human subjects dying from hypertensive complications had increased concentrations of cadmium or increased cadmium in relation to zinc.[1444] The systolic hypertension in rats caused by cadmium salts may be reversed by injecting a chelate of zinc with a higher stability constant for cadmium than for zinc.[430,1448] It has not only been concluded that cadmium plays some role in hypertension, but that the ratio of cadmium to zinc is very important. In metallothionein, which contains both zinc and cadmium, these two metals may interact with the same binding site and functionally replace each other. Since the physiologic role of this protein is not understood, these data cannot be specially related to renal pathology. All data suggest that, in the kidney, cadmium and zinc are closely related.[1451] In Figure 7-9, Schroeder *et al.* have shown that the concentration of zinc and cadmium in kidneys changes in amount and ratio in different decades of human life. The curves represent mean concentrations of 221 subjects. The difference in peak levels in age groups between 40 and 50 is statistically significant.

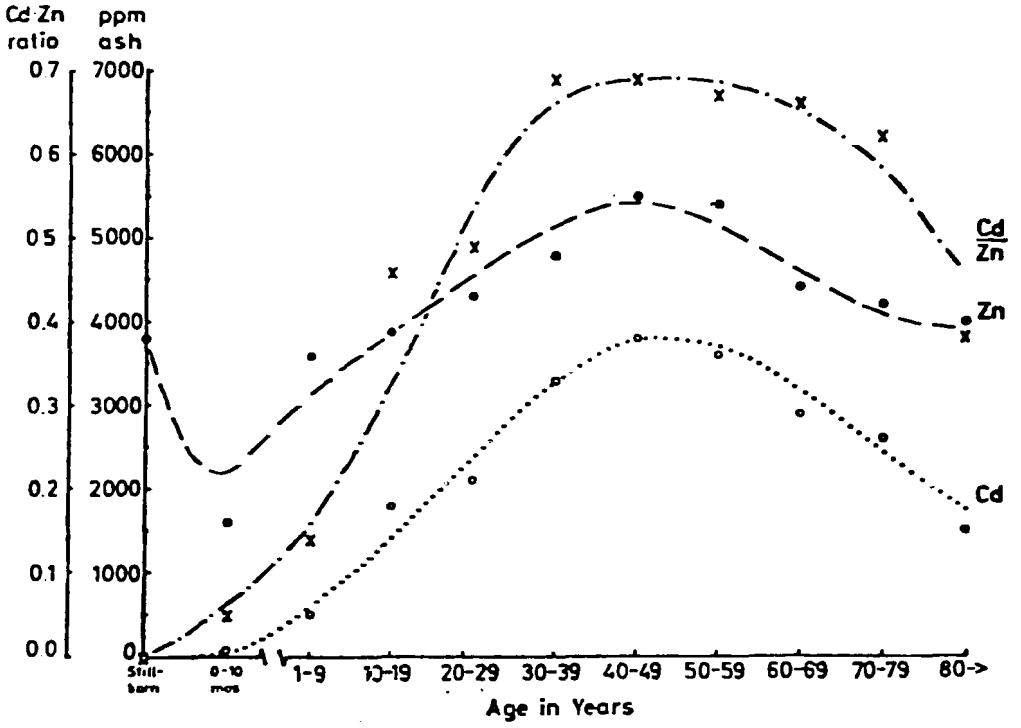


FIGURE 7-9 Mean concentrations of cadmium and zinc and their ratios in kidneys in 221 subjects from the United States. Reproduced from Schroeder et al.[1451]

Zinc has been found in a large number of urinary calculi, and in some patients with calcium oxalate renal calculi, hyperzincuria has been observed.[430,1336] Additional studies have suggested that only men with renal calculi exhibit hyperzincuria.[294,343] Hydrochlorothiazide has been suggested as a useful agent for preventing calcific renal stones.[1810,1811] Administration of this drug also produces a significant hyperzincuria,[1193] but evidently without significant cupruria. Zinc has been found to increase the solubility of calcium oxalate *in vitro*,[429] which may relate to the mechanism of this effect. However, sodium cellulose phosphate, another agent used in the treatment of some forms of renal stones, has no effect on renal excretion of zinc.[1192] Nephrocalcinosis was produced in rats fed a diet deficient in magnesium and zinc, and these renal stones were more common in females than in males.[1810]

Muscle Function

Several studies over the past 20 years have attempted to establish a role for zinc in muscle function. This function has not been clearly defined, but zinc might be necessary to muscle contractility. Although not directly related, examinations of cupric and zinc ions bound to crystalline sperm whale myoglobin and studied by X-ray diffraction analysis indicated that the metals are bound by histidine and probably by lysine and asparagine side chains with high affinity to myoglobin. Cann has suggested that zinc ion alters the iron-imidazole linkages in myoglobins.[227] The reaction of zinc with myoglobin causes major changes in physical and chemical properties of the protein. The most characteristic spectral change is a marked reduction of Soret band intensity, which shifts the peak of absorption to 390 mm.[226,227] Whether metal ions other than the iron atom of the heme group is physiologically involved in the function of myoglobin is not known.

Collagen

Recent studies in rats indicate an important role for zinc in collagen formation.[732] For example, a primarily copper- and iron-dependent enzyme, lysyl oxidase, was shown to be zinc-dependent as well.[733b] Cross linking *per se* appears to require zinc. Zinc deficiency appears to be associated with a decreased proline uptake in the epiphyses, a decreased transformation of proline to hydroxyproline, lowered sulfur-35 uptake and decreased epithelial shearing strength.[923,1142] Because zinc is important for the activity of alkaline phosphatase, and alkaline phosphatase has been related to the secretion of collagen and/or mucopolysaccharides, then a role for zinc in this system may be reasonably surmised. Zinc also may inhibit the pyrophosphatase activity of alkaline phosphatase in humans.[1083]

Hematopoiesis and Red Blood and White Blood Cell Function

In vitro studies of zinc-65 transport into leukocytes and erythrocytes and into erthyrocyte carbonic anhydrase have been carried out.[1041,1164] Zinc-65 uptake in leukemic leukocytes has been found to be only half that of normal cells.[377]

Carbonic anhydrase, first discovered in bovine erythrocytes, has been found in several human tissues.[831] Three forms have been found in human erythrocytes and named carbonic anhydrase A, B, and C.[33,901,1151] These different forms are similar in their role in catalyzing the reversible hydration of carbon dioxide, in their molecular weights, and in the one atom of zinc in each molecule.[301,1151] Studies of the mode of binding with different inhibitors, especially some derivatives of sulfonamides, described the mode of action of the active site and the differences between them.[302] Carbonic anhydrase B has at least one histidine molecule located in the active site region close to the metal.[1767,1768] Using zinc-65, it was shown by kinetics that immediate inactivation is caused by the formation of an inert ternary complex and that the mobilization of zinc proceeds slowly[235] by formation of the apoenzyme. Infrared spectrophotometry revealed that bound carbon dioxide attacked the hydroxyl coordinated to the zinc ion.[1346,1347] Other investigators studied the structure of the active site of this

enzyme; they constructed a model that corresponded to the active site.[388-390] Both human carbonic anhydrases B and C act as esterases on *o*- and *p*- nitrophenyl acetates and the mechanism of the carbon dioxide hydration reaction may be extended to the esterase reaction.[1684] Chelating agents such as *o*-phenanthroline at pH 4 and below may remove zinc and deactivate the enzyme.[414] The metal specificity of the enzyme seems to be independent of the substrate.[1613]

Alkaline phosphatase also has been found in white blood cells of man,[1365,1588,1629] and its concentration has been shown to be abnormal in several blood dyscrasias.[1365] Reverse transcriptase, a metalloprotein containing two zinc atoms per mole of protein,[1255] has been isolated and a function in human leukemia has been suggested.[596]

Sensory Function*

The role of zinc in taste and smell has been documented in humans and animals.[331,617,621,666,671,673,679,680,1004] Zinc deficiency is associated with loss of taste and smell acuity and the appearance of dysgeusia, dysosmia, and pica in man.[621,666,671] Zinc may be a component of the taste bud[666] and in subjects with normal taste acuity,[678,1087] it has been shown to be present in the saliva that bathes the taste bud. Several causes of loss of taste acuity in humans are associated with decreases in salivary zinc concentration[678,1087] and the appearance of pathologic changes in the taste buds.[671,680] Zinc is an integral part of a protein known to be secreted by the human parotid gland. The function of this protein appears to be related to the maintenance of normal taste bud form and function.[674]

Zinc also may affect the visual system. Zinc has been found in high concentrations in the choroid of the eye.[1655a] This concentration has been caused by

*See also Chapter 10.

localization of the metal in the tapetum lucidum cellulosum, which is thought to be responsible for the light reflection seen in the eyes of carnivores.[1754a] It is the reduction of the zinc content of this tissue that has been suspected to be the effect of chlorambucil in reducing the ophthalmologic reflection in canine tapetum lucidum.[1754a]

Retinene reductase, the alcohol dehydrogenase which converts retinol to retinal, is a zinc-dependent enzyme. The presence of this enzyme in the retina partly may explain the high level of zinc in the retina.[126] This enzyme may also affect the olfactory system because vitamin A levels and hyposmia are associated in several pathologic states.[672] Zinc levels in the olfactory epithelium have not been measured yet.

Liver

Several zinc-dependent enzymes have been found in liver. Liver alcohol dehydrogenases catalyze the same reaction as in the retina, that is, a reversible oxidation and reduction of alcohols to the corresponding aldehydes or ketones in the presence of nicotinamide adenine dinucleotide (NAD) or reduced nicotinamide adenine nucleotide (NADH). They have different amino acid compositions and different total and essential sulfhydryl groups.[636] An early study reported that horse liver alcohol dehydrogenase had two zinc atoms per mole of enzyme,[1607] but in later work using[a] zinc-65 chloride tracer[13] as well as nonradioactive methods,[1173] four atoms of zinc per mole were identified in this protein.

Zinc porphyrins are commonly found in liver. Zinc protoporphyrins have been found in yeast[1161] and zinc chelatase, an enzyme that assists in incorporating zinc into protoporphyrin has been detected in different extracts.[1134,1292] Zinc ferrochelatase also may act to incorporate zinc into protoporphyrins.[888] There is also some support for the notion that zinc protoporphyrin and iron porphyrins are catalyzed by the same enzyme, as both enzymes

in the native state are closely associated with phospholipids.[998] In *Saccharomyces cerevisiae* several zinc porphyrins have been isolated, suggesting that zinc protoporphyrins are present in intact cells.[1292]

Pancreas

Carboxypeptidase has been purified from bovine and porcine pancreas, with small variations in properties. Bovine pancreatic carboxypeptidase A has α-, β-, γ-, and δ- forms that differ in solubility with respect to the amino acids of the N-terminal residue[884] and in reactivation of the apoenzyme by zinc. The amino acid composition for most types has been determined.[76,1791] The zinc atom of the active site of γ- and δ- forms is bound to the thiol group of the cysteine residue[1248] and the α-amino group of the N-terminal of asparagine.[313] In carboxypeptidase B, one cysteinyl residue is the ligand for zinc.[1793] Many studies have been conducted recently to evaluate the active site and investigate relationships between structure and function.[121,305,306] Carboxypeptidase B activity is decreased in zinc-deficient rats.[1066]

Vitamins

Hsu reported that pyridoxine-deficient rats have low zinc content in plasma, liver, heart, and pancreas.[728a] Conversely, Gershoff found that pyridoxine and vitamin B_1 deficiencies increased the zinc content of rat pancreas, liver, and kidney. The greatest increase in zinc was in the content of the pancreas. In pyridoxine-deficient rats the decreased availability of insulin was postulated as the cause of the higher zinc levels. *In vitro* studies with adipose tissue isolated from these rats indicated that zinc inhibited the lipogenic activity of the adipose tissue, suggesting that it might influence the control mechanisms related to insulin activity.[387]

A role for zinc in vitamin A metabolism has also been suggested,[1512] but the relationship is unclear because of the complex interactions between zinc and protein metabolism and between zinc and vitamin A. This proposed relation may be more related to the availability of protein than to vitamin A itself--[1505] were observed to anorexic, zinc-deficient animals/take in less than normal amounts of protein.

Cardiovascular System

Little is known about the specific role of zinc in the function of any particular aspect of the cardiovascular system, but important relationships have been postulated. As already noted, an interaction between renal cadmium and zinc has been suggested as an influence on some forms of hypertension.[1444,1448] The urinary excretion of cadmium was greater in 95 hypertensive women treated with antihypertensive agents than in 107 normotensive women, but zinc excretion was the same in both groups.[1013] Carroll attempted to link the zinc concentration in air and the incidence of cardiovascular disease--including hypertension--in 28 North American cities.[240] His results are difficult to correlate with the multiple / clinical factors that directly or indirectly relate to the pathogenesis of the disease processes.

Some investigators have suggested that zinc deficiency may be a factor in the development of atherosclerosis[686,1261] and that zinc therapy is useful in the treatment of some of the symptoms of obliterative vascular disease.[686] One investigator noted that the zinc content of the aorta of atherosclerotic patients, particularly in the areas of the atherosclerotic plaque, was markedly decreased;[1703] he also noted that these patients exhibited decreased zinc in plasma, liver, myocardium, pancreas and hair. It has been suggested that accumulation of zinc in the renal cortex promotes aortic atheromata and hypertension.[1133] Another group experimenting with hypertensive dogs could not find these large differences;[474] these studies were carried out in animals with a different set of cardiovascular problems. Zinc-protamine-glucagon has been claimed to be useful

in the relief of symptoms related to cardiac insufficiency,[812] although the role of zinc itself in this complex may not be critical.

After acute myocardial infarction, human serum zinc concentration falls.[934,1711] Dogs with experimentally produced myocardial infarctions had reduced zinc concentrations in the infarcted area.[936] However, no differences in zinc were found in heart muscle analyzed from patients who died from acute coronary thromboses although magnesium levels were decreased.[274] In addition, zinc was carefully measured in the thoracic aorta of patients with graded levels of atherosclerosis and differences between those subjects and patients not suffering from cardiovascular disorders could not be measured.[1310] Such diverse results make it difficult to ascertain the specific influence of zinc in cardiovascular function.

Other Enzymatic Activities

Zinc complexes with adenosine diphosphate (ADP) to form Zn_2ADP, ZnADP, and $Zn(ADP)_2$ in dilute solutions. Zinc markedly reduced the conversion of adenosine triphosphate (ATP) to AMP in lipocytes when used alone or with magnesium.[1784] ATP-metal-norepinephrine ternary complexes may be of major importance to the understanding of the biologic phenomenon of binding or storage of catecholamines *in vivo*.[298]

Zinc has also been found to affect the activity of ATPase,[244,1231] acid phosphatase,[1728] alkaline β-glycerophosphatase,[287] phosphopyruvate hydratase, and yeast aldolase.[1091] Zinc inhibits the activity of several enzymes, including ribonuclease,[158,690] allantoinases from bacterial and animal sources,[1698] α-oxoglutarate dehydrogenase,[1742] and sodium- and potassium-activated ATPase.[1272] The character of the inhibition for each enzyme is specific.

Superoxide dismutase, an enzyme important in oxygen metabolism,[1005] contains two zinc atoms, perhaps as structural components of the enzyme.[645] Zinc is bound at the histidine site of this enzyme.[490]

Zinc-α_2-glycoprotein, with a molecular weight of 43,000, is present in most body fluids and has been isolated from normal plasma.[1427] It is found in low concentrations in normal serum and cerebrospinal fluid, and in higher concentrations in urine, saliva and sweat.[1260] This glycoprotein appears to be homogeneous, as shown by ultracentrifugation, electrophoresis, chromatography and N-terminal-amino acid analyses. In the low pH range, the protein appears in several fractions even after the removal of sialic acid. The polymorphism of zinc-α_2-glycoprotein may be an expression of genetically determined variants.[1429] The zinc-α_2-glycoprotein should not be confused with the major serum macromolecular binding protein of zinc-α_2-macroglobulin noted in the beginning of this chapter.

CHAPTER 8

ZINC IN THE DIET AND THE EFFECTS OF ZINC DEFICIENCY IN ANIMALS*

Zinc must be present in the diet of all animals, including man. Moreover, it must be supplied in the diet almost continuously, because animals have only small amounts of readily available zinc stored in the body. This condition exists although the body contains a relatively large amount of zinc, particularly in bones, skin, and hair.[1640] However, zinc in these tissues cannot normally be utilized by the animal to meet its need for zinc.

LACK OF ZINC IN THE DIET

The primary effect of insufficient zinc in the diet is decreased food intake and cessation of growth. The effect is quite rapid; growth stops almost immediately in rats[1195,1782] and in about 2 wk in calves and lambs.[1064] Other signs of zinc deficiency observed in young growing animals are listed in Table 8-1.† It should be emphasized that the additional symptoms described generally were observed in very young animals that would have been in an active stage of growth. Somewhat older animals will also stop growing soon after being placed on a zinc-deficient diet but may not show other signs of deficiency. Six-to-nine-month old calves, for example, failed to grow after being placed on the deficient diet, but showed no other clinical signs of deficiency.[1064]

Undoubtedly, adult animals are also affected by a lack of zinc in the diet, but less information exists on the effects of a low zinc diet

*Work discussed in this chapter appeared in the literature prior to April 1975.[1640] Papers included in Underwood's comprehensive review generally have not been covered here.

†See Chapter 10 for symptoms in humans.

TABLE 8-1

Symptoms of Zinc Deficiency in Young Animals and Approximate Requirements for Zinc

Species	Deficiency Symptoms[a]	Approximate Requirement[b]
Ruminants		
Cattle	Parakeratosis, increased bacteria in mouth, stiffness of joints with swelling of feet, horny overgrowths, excessive salivation (transitory)[1050]	Egg white: 10-14[1064] Practical diet Beef cattle: 10-30[1114a] Dairy cattle: 40[1108]
Sheep	Salivation (transitory), increased bacteria in mouth, parakeratosis of tongue,[973] loosening of wool, distortion of hoof walls and horns[1064]	Egg white: 15[1064] (somewhat higher level may be required for normal testicular development)[1642]
Goats	Similar to cattle[1127]	
Swine	Parakeratosis[1114]	Casein diet: 14-20[1479] Soy diet: 50[1114]
Poultry		
Chicken	Poor feathering, shortening and thickening of long bones of legs and wings, hock enlargement, reduced egg production and hatchability, skeletal abnormalities in embryos[1113]	Casein diet: 12[1156] Soy diet: 19[1156] Practical diet[1113] Starting chick (0-8 wk):50 (1.5 times this amount may be required during first week to prevent abnormal feather development)[1572] Breeding hens: 65
Turkey		Poults (0-8 wk): 70[1113]
Japanese quail	Abnormal feathering, uncoordinated gait[1699]	25[1699]
Laboratory Animals		
Rat	Alopecia, dermal lesions, parakeratosis of the esophagus,[383] cyclic food intake,[1782] impairment of reproductive function[1640]	Egg white or casein diet: 12-13[846,1112,1197,1782] Soy diet: 19[1156]

TABLE 8-1 continued

Species	Deficiency Symptoms	Approximate Requirement
Mouse	Alopecia[1112]	>3[1112]
Rabbit	Sparse hair, reddening around mouth, wet and matted hair on lower jaw and ruff,[44] impairment of reproductive function in the female[44,1480]	>3[44]
Guinea pig	Rough coat, scaly dermatitis[1001]	Casein diet: 12 Soy diet: 20[15]
Monkey	Unkempt appearance, alopecia, parakeratosis of tongue[79]	Casein diet: ≥15[961]
Dog	Alopecia, hyperkeratinization, acanthosis[1109] (symptoms and requirement based on single study in which a simple zinc deficiency probably was not present)	Practical diet: 20[1109]
Mink		≤20[c]

[a] Symptoms listed are in addition to growth failure and decreased food consumption.

[b] Mg/kg feed, dry weight.

[c] Unpublished data, H. F. Travis.

on adult animals. Studies with rats indicate that a low zinc diet interferes with wound healing in the adult male [1154,1408] and with estrous cycling [45,544] and reproduction in the adult female. [1640] Skin lesions have been reported in adult humans whose zinc intake has been very low for 2 mo. [199]

REQUIREMENT FOR ZINC IN THE DIET

Table 8-1 also estimates the amount of zinc that various animals require in order to grow normally. Adult animals presumably require somewhat less than the amounts listed, except during periods of stress such as pregnancy and lactation. The amount of zinc required varies somewhat with the type of diet, because diets which contain seed protein (such as soy protein) may increase the requirement for zinc. There may be other conditions under which the requirement for zinc is increased, since deficiencies have been reported in ruminants in the field on zinc intakes which, based on laboratory studies, should have been adequate.

A lower than normal requirement for zinc has been reported in germ-free rats [1513] and a higher than normal requirement in a breed of Danish calves with hereditary thymus hypoplasia. [179] A genetic difference has been suggested to affect the requirement of pigs.[898]

Normally, little zinc is available to the body except that ingested in the diet, and therefore zinc must be supplied in the diet more or less continuously. Under some conditions, however, zinc stored in the tissues may become available to the body. Such conditions include muscle and/or skin catabolism after severe injury, [462] starvation, [1532] low protein intakes, [760] and bone resorption during low calcium intake. [762,1599] The animal may also be able to make some adjustment to a low zinc intake

through decreased excretion of zinc and perhaps by increased absorption [199,447,1048,1456] efficiency.

Ruminants

Cattle. The suggested zinc requirement for cattle is 40 ppm for dairy cattle [1108] and 10-30 ppm for beef cattle. [1114a] This rather wide range exists because zinc deficiencies occur in the field on intakes of zinc substantially higher than those which appear to be adequate under experimental conditions.

Under experimental conditions, 8 ppm was sufficient for calves to grow, although plasma zinc levels were still low. [1064] This concentration may be very close to the minimum, because in another study calves receiving 7-8 ppm became severely deficient. [1052] On an intake of 17 ppm, lactating cows did not show signs of deficiency nor was milk production affected, although the zinc concentration of the milk was decreased by about 25%. [1123] Cows receiving low levels (5-17 ppm) of zinc absorbed and retained [1124,1126,1212,1213] more zinc-65 than did cows fed 40 ppm. [1125] Addition of zinc to a barley-hay ration containing 25-32 ppm zinc did not improve weight gain of beef heifers. [1302] The experiments indicate that 30 ppm should be an adequate zinc intake for beef cattle; but zinc supplements given to beef calves in Greece on a ration already containing 32 ppm zinc resulted in a 7% greater weight gain, and deficiences were reported in the field on intakes of 20-40 ppm. [1528] Young Fresian bulls on a barley-swede turnip ration containing 30-50 ppm zinc developed a pododermatitis that was healed by oral zinc administration [375] although growth was never affected. Facial eczema in dairy cattle in New Zealand was treated successfully with zinc supplements, [1345a] although forage in the area had not been considered to be low in the metal. [552]

The above cases suggest that some unknown factor influences the zinc requirement of cattle. One possibility is phytate, although phytate content of the diet has not been considered to be a problem in ruminants.[1050] Another factor that has not been carefully considered is soil ingestion. Dairy cows may consume 1 lb (0.45 kg) or more of soil a day.[657] In some cases, animals might be able to utilize the zinc in the soil,[658,1510] but ingestion of other soil types might reduce the availability of zinc. Stress also may increase the requirement for zinc.

Sheep and goats. Although severe zinc deficiency has been developed in sheep,[1064,1204] little evidence exists of zinc deficiency occurring in sheep in the field. A field survey of plasma levels of zinc in sheep in various parts of New Zealand[552] indicated that sheep in some areas had plasma zinc levels as low as animals reduced in growth by experimental zinc deficiency.[1064,1204] However, no cases of zinc deficiency were observed. A survey of 600 sheep in Greece showed that 1% of them had severe symptoms of zinc deficiency, and 60% had milder symptoms probably associated with other factors (other deficiencies, poor management, etc.).[1204] Zinc or zinc plus manganese supplements were reported to have improved reproductive performance in ewes in Australia, although management was apparently also involved.[418]

It is not known whether sheep are more resistant to zinc deficiency than cattle, or if zinc deficiency in the field merely has not been recognized. Sheep may need less zinc than cattle because deficiencies developed in sheep on a dietary level of 4 ppm zinc[1204] in the same amount of time in which deficiency symptoms developed in calves on 7-8 ppm dietary zinc.[1052] The large amounts of soil ingested by sheep[656] and the ability of sheep ruminal fluid to extract zinc from it[658] may also offer protection.

Zinc deficiency in goats has not been studied much. Nine-month-old male goats on a diet of 4 ppm zinc developed signs of deficiency in about 5 wk.[1127] Adult female goats on a diet of 6-7 ppm did not develop signs of zinc deficiency until they were lactating. Milk production was not affected, but the zinc content of the milk dropped to approximately 50% of that produced by the controls after a month's lactation.[569] Male kids raised on the 6-7 ppm diet were severely stunted, but females apparently were not affected.[375,945] Other observations have indicated that the growing intact males have an increased zinc requirement, but the differences have not been as large as those reported in this experiment. It would seem that other factors must have been involved.[1204] The survey of ruminants in Greece mentioned previously found that 2% of the 150 goats examined exhibited severe symptoms of zinc deficiency.

Swine

Although 14-20 ppm zinc appears to be adequate for pigs on a diet with casein as the protein source,[1479] the requirement on a soy-based diet (in which the zinc is less available) is 50 ppm.[1114] However, soy is not the only feed that increases the zinc requirement; pigs on a cassava-rice bran diet supplying 40 ppm zinc displayed symptoms of zinc deficiency, whereas pigs on a corn-soy (20%) diet with 48 ppm zinc developed normally.[992] Swine receiving a 13% soy meal intake showed no signs of deficiency although the zinc concentration was only 32-34 ppm. Not surprisingly, neither the additions of ethylenediaminetetraacetic acid (EDTA)[1188] nor pre-chelated trace minerals[1187] to this diet improved weight gain of the pigs. Addition of low levels of the pre-chelated trace minerals was said to produce poorer results than the basal diet without the added trace elements;

but none of the results were strikingly different and considerable discrepancy was found between the amount of trace minerals calculated to be in the diet and the actual content determined by analysis.

For the pig in particular, high calcium levels increase the zinc requirement in diets containing large amounts of seed protein.[614]

Rat

Zinc deficiency has been studied extensively in the growing rat.[99,846,847,1195,1196,1197,1236,1640,1782] A casein diet containing 8 ppm zinc yielded maximum growth, but 15 ppm was required to maintain bone and whole body zinc at normal levels.[1197]

Humans

Zinc was included in the list of recommended dietary allowances (RDA) for humans for the first time in 1974,[1119] and recommended levels are given in Table 8-2. These recommendations are based partly on the average intake of apparently healthy people and partly on balance studies. In a balance study the amount of zinc ingested in the diet is compared to that excreted in the urine and feces. Because the studies are cumbersome, they usually involve small numbers of people for short periods of time on a limited range of intakes. Such data allow only a crude approximation of the requirement, but they provide the best information currently available. Levels similar to those of the RDA's also were arrived at by a committee of the World Health Organization.[1806] Requirements in this case were based partly on considerations of changes in lean body mass at various ages.[1404]

The adequacy of 15 mg zinc daily for adults is supported by recent studies in which both adult American men[1532] and young New Zealand women[1354]

TABLE 8-2

Recommended Dietary Allowances For Zinc[a]

	Age	Weight		Height		Zinc
	(years)	(kg)	(lbs)	(cm)	(in)	(mg)
Infants	0.0-0.5	6	14	60	24	3
	0.5-1.0	9	20	71	28	5
Children	1-3	13	28	86	34	10
	4-6	20	44	110	44	10
	7-10	30	66	135	54	10
Men	11-14	44	97	158	63	15
	15-18	61	134	172	69	15
	19-22	67	147	172	69	15
	23-50	70	154	172	69	15
	51+	70	154	172	69	15
Women	11-14	44	97	155	62	15
	15-18	54	119	162	65	15
	19-22	58	128	162	65	15
	23-50	58	128	162	65	15
	51+	58	128	162	65	15
Pregnant						20
Lactating						25

[a]From Recommended Dietary Allowances.[1119]

were in balance on a daily intake of approximately 17 mg. Adults have also been reported to be in balance on lower intakes.[504,614,643,1119] A zinc intake of 4.5 mg in a diet typical of low income groups appeared to be marginally adequate for 7-9-yr-old girls when additional nitrogen was added to the diet.[1298] Levels of zinc in diets of children from different institutions varied from 2.7-6.4 mg/day.[1102] The allowance for the infant is based primarily on the estimated zinc intake of an infant receiving breast milk. This amount may not be enough to maintain bottle-fed infants in positive zinc balance, because an intake of 1 mg/kg body weight was necessary to keep infants less than a month old in positive zinc balance. In 3-4-month-old infants, positive balances could be maintained with 0.75 mg.[471] The rapid decline in the zinc concentration in infants' hair in the U.S. shortly after birth[617] may also be an indicator that zinc intake is marginal. Hair zinc in infants in Thailand was not low after birth,[615,621a] and hair zinc in infants in England did not decline as much as it did in the U.S.[614a] In addition, supplementing an infant milk formula with zinc increased the growth of male infants by age 6 mo.[1722] However, evidence from animal studies shows that some change in zinc metabolism takes place after birth, as plasma zinc levels decreased shortly after birth in both rats[757] and pigs.[879] More information is needed on the zinc requirements of infants and children.

Total parenteral nutrition[562] also substantially lowered the plasma zinc levels in two infants. Trace element formulas for use in intravenous hyperalimentation solutions for children and adults have been described.[749,1483a]

Reproduction

Female. Zinc requirements for pregnancy and lactation have not been extensively studied. In the rat and pig, the zinc level that allowed maximum growth was also adequate for gestation: 12 ppm in the rat [1780,1782] and 20 ppm for swine on a casein diet and 34 ppm for a corn-soy diet. [65,1607b] The requirement for lactation is assumed to be higher than that for gestation. An adequate zinc intake for gestation in the goat resulted in severe deficiency during lactation. [569]

Little is known about the effect of zinc on reproduction in the human female except that plasma and hair zinc levels decrease during pregnancy [616] and with intake of oral contraceptives. [614] To some extent, this may simply reflect a redistribution of zinc in the body. [1000] Pregnant teenagers who were given 30 mg zinc daily still maintained lower levels of plasma zinc during the ninth month of gestation than they had 6 wk after parturition. [1439] Nonetheless, the values were higher than those reported for normally nournished women in the ninth month of pregnancy. [616] Zinc concentrations in plasma and hair two days after parturition were lower in women in Iran that had been living in villages or under low socioeconomic conditions in the city than in those that had been living in higher socioeconomic levels in the city. [1414] (But an attempt to correlate decreased plasma zinc with abnormal outcome of pregnancy in East Harlem was unsuccessful. [1515]) Several unsuccessful pregnancies have been reported in patients with acrodermatitis enteropathica, [618] a condition in which very low plasma zinc levels have been reported. It recently has been discovered that the condition responds to zinc therapy. [1085,1129a,1520a]

The possibility of zinc deficiency occurring in human pregnancy has been of particular concern [721,1474a,1475] because zinc deficiency in rats has been shown to cause congenital malformations. [757,759, 761,761a,1640,1732] To what extent these results are applicable to humans is

unknown. When evaluating these results one must remember that extremely low zinc diets are required to produce the malformations, and that so far the only reported experiments have been conducted with rats. The results of the rat studies may not be applicable to other species since vitamin deficiencies, for example, have been known for a long time to be teratogenic in the rat; but comparable effects have not been seen in humans.[1730] Similar experiments are difficult to simulate in other animals, and apparently no experiments have been done on animals with long gestation times and only one or two offspring. For animals that do not bear large litters, zinc may be more important to normal mating and maintenance of pregnancy.

Male. A zinc intake somewhat higher than that required for normal growth may be necessary to support normal reproductive function in the male. Male sheep receiving a diet containing 17 ppm zinc exhibited reduced testicular weight, low volume of seminal fluid, and an increased percentage of abnormal sperm, compared to sheep receiving 32 ppm zinc, although food intake and weight gains for the two groups were the same.[1642] Young bulls developed a zinc-responsive pododermatitis, although steers and heifers were unaffected.[375]

In the rat, the developing testes seem to be particularly susceptible to zinc deficiency.[383,1174,1640,1642] Because the testes of rats with restricted food intake are not affected, the effect appears to be primarily a result of lack of zinc and not secondary to low food intake.[383,1642] Changes in the adult male goat after relatively longer periods of zinc deficiency were apparently related more to lowered food consumption.[1127]

SOURCES OF ZINC IN THE DIET

In an average diet, most of the zinc will be supplied with the protein ingested. Table 8-3 sets forth the approximate zinc content of foods relatively high in zinc.

TABLE 8-3

Zinc Content of Selected Human Food

	mg/100 g (3.5 oz serving)
Oysters	100[a]
Roast beef	4.0-8.0[b]
Chicken, dark meat	3.0[b]
white meat	1.0[b]
Fish (perch)	2.5[b]
Liver	4.0[b]
Egg	1.5 (0.7 mg/egg)[a]
Nuts	3.0[a]
Wheat germ	13.0[a]
Whole wheat bread	3.0 (1 mg/slice)[c]
White bread	1.0[c]
Legumes	1.0-5.0[a]
Milk	0.3/100 ml (1.4 mg/0.47 l)[b]

[a] Data from Schlettwein-Gsell and Mommsen-Straub.[1425]

[b] Data from Osis et al.[1179]

[c] Data from Zook et al.[1826]

One serving of oysters will more than meet the adult zinc requirement for the day. Two servings of meat will probably supply approximately half the daily requirement. Nuts, legumes, and whole grains also contain relatively large amounts of zinc, but the zinc in these foods may not be as available as the zinc in meats (see discussion of phytate).

The zinc content of foods as published in the literature has been comprehensively reported.[1425] Some additional zinc values of various foods are listed by Osis;[1179] of wheat and wheat products by Zook;[1826] and of market milk and evaporated milk, infant products, and human milk by Murphy.[1100, 1101] Zinc contents of a few forages and animal foods have also been reported.[1115] Representative zinc values for a number of common foods have been compiled by the Consumer and Food Economics Institute, United States Department of Agriculture (USDA).[1093a] These tables listing zinc content of foods make up Appendix A.

Since the values for cooked meat and fish[1179] are not very different from what would be expected based on the values given for raw muscle, loss of zinc in cooking meat appears small. Canned vegetables have been reported to have 40-80% less zinc than fresh vegetables.[1446] Since a negligible proportion of zinc in the diet comes from fruits and vegetables other than legumes, loss of zinc from this source would be of little consequence. In addition, the canned and fresh vegetables that were compared in the study were not from the same lot, so zinc contents may have been different even before processing. The most serious losses in processing are those that occur in refined foods such as wheat. For example, whole wheat bread has three times as much zinc as white bread.[1826]

Although the concentration of zinc in milk is relatively low, an adult who drinks a pint (0.47 l) of milk a day can get 10% of the zinc requirement from this source. Reports of zinc concentration in cows' milk varied from 3 to 8 μg/g,[1101,1593] with 4 μg/g given as the average.[1093a] Human milk was reported to have the same[1771a] or less[1100,1175,1244] zinc than cows' milk. It is possible that zinc in human milk is absorbed more efficiently, however, since plasma zinc levels fell from 60 to 39 μg/100 ml in a 2-yr-old boy with acrodermatitis enteropathica when he was no longer given human milk.[619] Improvement in patients with acrodermatitis who are given human milk[1085] may, of course, be from some factor other than increased zinc absorption. Colostrum[1107] and partum mare's milk[1639] had twice as much zinc as milk obtained at later times.

Availability

As was mentioned when discussing the requirement for zinc, the amount of zinc needed in some diets is higher than in others. Presumably this is caused by a difference in the availability of zinc in these diets. (Some factors associated with reduced availability of zinc in certain diets are discussed in the following section on interrelationships between zinc and other dietary components.) Some work has been done to determine the best way of measuring the availability of zinc.

Differences in zinc concentration that varied with protein source were found in tibia of young Japanese quail, although the birds' body weight was not affected.[634a] Rats on a moderately low zinc diet also showed a difference in tibia zinc, although again body weight was similar.[104] Tibia zinc increased in rats from 3-16 weeks of age on normal diets.[103] Total zinc in the femurs of young rats repleted with 12 ppm dietary zinc

were reported to be linear.[1076] In a similar experiment, however, the zinc content of bone increased only slightly.[1371] Bone zinc concentration decreased rapidly in rats on a low zinc diet.[254,1371] In rats, at least, bone zinc does not seem to be any better a measure of zinc availability than is an increase in body weight in rats fed diets in which the amount of zinc is less than that required for normal growth.

Fortification

To meet the RDA of 15 mg zinc/day, one must eat a fairly high protein diet. From the data in Table 8-3, one can estimate that an adult who has an egg and two pieces of whole wheat toast for breakfast, chicken for lunch and roast beef for supper would consume approximately 12 mg zinc. If meat is eaten only once a day, with cereals or legumes as the protein source for other meals, the zinc intake would be significantly less. To raise the zinc content of low protein diets, fortification of cereal foods with 22 ppm zinc has been recommended by the Food and Nutrition Board of the National Research Council.[1117] Even with fortification of cereals it would be very difficult for a woman on a low protein diet to meet the recommended allowance of zinc for pregnancy and lactation. Even a woman on a high protein diet would have difficulty meeting the 25 mg requirement for lactation without a zinc supplement.

Institutional diets generally fall below the 15 mg/day standard for zinc.[1033,1451]

INTERRELATIONS BETWEEN ZINC AND OTHER DIETARY COMPONENTS

Phytate

Since most instances of zinc deficiency in the field have occurred in diets in which most of the protein was obtained from plant sources

(and seeds in particular), zinc from plants is considered to be less available to monogastric animals and chickens than is zinc derived from animal protein. This distinction has been attributed to the presence of large amounts of phytic acid (inositolhexaphosphoric acid) or phytate in seeds. It has been recognized that soy protein increases the need for zinc in the diet, presumably because of the protein's phytate content.[904] Other seed meals have the same properties as soy so that their use also might increase the zinc requirement. Protein concentrate from mustard or rapeseed which also contained high levels of phytate interfered with reproduction in female rats when it was fed from the time of breeding to parturition.[1015a] Since serum zinc levels were low, the effects were attributed to zinc deficiency; but the performance of females receiving these diets along with increased levels of zinc was not tested. A cassava-rice bran diet caused zinc deficiency in pigs, although the zinc level in the diet was 45 ppm.[992] The high level of phytate in rice bran--approximately 5%--may have been responsible for the high zinc requirement. (Phytate contents of some feed ingredients are supplied by Gontzea and Sutzescu[545] and Nelson *et al.*[1132])

The incidence of zinc deficiency in Iranian villagers was attributed to high levels of phytate in the bread.[1325] To demonstrate the effect of phytate, high levels of phytate in the chemical form or *tanok*, an unleavened bread high in naturally occurring phytate, were fed to three men. Zinc excretion increased in two of the three men, but serum zinc levels were not consistently lowered.[1328] Prasad *et al.* reported that Iranian men whose diet consisted almost exclusively of bread and beans had low levels of zinc in their plasma, red blood cells, and hair, suggestive of zinc deficiency.[1276]

Incubating zinc-65-labeled high extraction wheat with yeast increased the solubility of zinc-65 in the wheat.[1329]

Since phytate phosphorus decreased in yeast-wheat mixtures,[1326] part of the increased availability was probably from hydrolysis of phytate. However, the difference in solubility of zinc-65 was much greater than could be accounted for by phytate destruction alone. There are also enzymes in the animal intestine that degrade phytate,[122,361a,1778] but they have not been studied extensively.

Adding phytate to animal proteins decreased the availability of their zinc to chicks; the effect was aggravated in the presence of high levels of dietary calcium.[1155] Assay of zinc availability in plant food ranged from 40-60%,[1156] quite a high availability compared to that of other minerals. Zinc in zinc-65-labeled pea seeds was 75% available even in mature seeds with a 1.2% phytate content.[1755] Hence, factors other than phytate contribute to the appearance of zinc deficiency in animals on diets high in plant protein. The high fiber content of these diets may also contribute to reduced zinc availability since fecal zinc loss has been reported to be significantly correlated with fecal dry solids.[1439]

The presence of phytate in the diet has not been considered a problem in ruminants because the microorganisms in the rumen can degrade phytate.[545,1050] The development of a zinc-responsive pododermatitis in young bulls on a diet containing swede turnip and barley[375] suggests that this conclusion should be reexamined, however, since the phytate content of this ration could be relatively high.[545] Weight gain of cattle on a soybean meal ration was improved by adding a combination of trace elements, including zinc, to the ration.[288]

Calcium

Calcium generally does not depress zinc absorption except in the presence of phytate.[1155] Addition of 7.7 g calcium lactate

to the diet of 7-9-yr-old girls, for example, did not change the excretion of zinc,[1298] nor did a sixfold increase in the calcium intake (from 200 to 1,300 mg) affect the zinc balance in adult men.[1532] Phytate reduced the rise in serum zinc in normal adults after zinc sulfate dosage, but calcium had no additional effect.[1216a] However, some indication exists that high levels of calcium may affect zinc absorption when zinc levels in the diet are marginal. Rats given 1.3% calcium excreted more zinc-65 in the feces than did controls given 0.6% calcium. The effect appeared to be at the intestinal level, since the calcium level in the diet had no effect on retention or distribution of zinc-65 injected intraperitoneally.[741] The high level of calcium (18%) in the clay ingested by some Iranians[1510] may also have contributed to zinc deficiency in this population. Because the source of much dietary calcium in this country is milk, it is interesting that oral administration of lactose and zinc-65 increased the absorption and retention of zinc-65 in rats.[500]

A low calcium diet has been reported to reduce the severity of zinc deficiency in the pregnant rat, presumably by causing resorption of bone and thereby releasing zinc from it.[762] To support this theory, a low calcium diet was shown not to alleviate zinc deficiency in pregnant rats parathyroidectomized to prevent bone resorption.[1599] Pigs on a low zinc and low calcium diet showed no signs of zinc deficiency, although those on a high calcium-low zinc diet were severely affected.[1146] Bone zinc was lower in pigs on the low calcium-low zinc diet, suggesting that either mobilization of zinc from the bone or failure to deposit zinc in the bone had occurred. In growing rats on a low calcium diet with a normal amount of zinc, bone zinc increased.[464]

Protein and Amino Acids

Net protein utilization of a soy diet by chicks was increased slightly when zinc was increased from a marginal to an adequate level.[57] Low levels of dietary protein in rats decreased both zinc absorption and levels of zinc in the liver and small intestine,[1671] implying that a secondary zinc deficiency might be induced in cases of protein malnutrition. Zinc absorption and retention by ruminants similarly was decreased on a low protein diet.[1543] In the ruminant, the effect was attributed to a lessened need for zinc because of reduced growth, but the lowered tissue zinc concentrations found in the rats on the low protein diet suggests interference in zinc metabolism.

Addition of 1% histidine or 0.2% histamine to low zinc diets alleviated leg abnormalities in chicks[715] and skin lesions in pigs[349] without affecting the zinc concentration of the tissues.[1324] Histidine did increase alkaline phosphatase in tibia of chicks fed a low zinc-soy protein diet.[905] Feeding a histidine-rich protein to pigs also alleviated zinc deficiency, although here absorption of zinc was increased.[350] Adding arginine to chick diets low in zinc increased the severity of the leg abnormalities. Even with adequate zinc, added arginine tended to depress tibia zinc concentration, though not as markedly as with low levels of zinc.[300] Since many seed proteins are high in arginine, arginine, as well as phytate, may cause problems when seed proteins are used in chick diets.[904] The negative effects may be peculiar to chicks since their metabolism of arginine differs from that of mammals. Unlike the experimental animals, humans fed a diet rich in histidine became zinc-depleted.[673]

Vitamins

Biotin has also been reported to alleviate somewhat the symptoms of zinc deficiency in the rat.[282] However, the observation was based on a small sample of rats. In a more extensive study, biotin levels from 2-50 mg/kg diet had no effect on weight gains or zinc levels in rats on a low zinc diet.[1201] Since folic acid concentration has been reported to be low in livers of zinc-deficient animals,[1781] these workers also studied the effect of adding 1-5 mg folic acid/kg diet. The additional folic acid produced no detectable effect. Similarly, additions of 2, 5, and 10 times the requirement for thiamine, niacin, riboflavin, pyridoxine, pantothenic acid, or vitamin B_{12} failed to alleviate any of the symptoms of zinc deficiency.[1200]

Low levels of vitamin A have been reported in zinc-deficient lambs[52,1413] and rats.[383a,1512] Retinol-binding protein was also lower in serum of zinc-deficient animals.[1514] Since vitamin A levels in the livers of zinc-deficient rats were higher than in *ad libitum* controls,[1512] it was concluded that zinc is required for normal mobilization of vitamin A from the liver. The accumulation of vitamin A in the liver of the deficient rat was related to the animal's lack of growth, however, because zinc-supplemented animals with growth restricted by limited food intake to the same rate of growth as the zinc-deficient animals also had high levels of vitamin A in the liver and low levels in serum.[1511] The low serum level in lambs may also have resulted from slow growth. Low levels of vitamin A in serum have been observed in protein-calorie malnutrition.[1356]

Other Minerals

Since cobalt can replace zinc *in vitro* in some zinc-dependent enzymes,[929] the possibility of substituting cobalt for some of the *in vivo* zinc requirement has been investigated. A preliminary report indicated that this substitution was possible in pigs.[716] In a more recent study with rats, cobalt reduced the zinc content of liver and serum but was ineffective in relieving any zinc deficiency symptoms. In the rat, cobalt cannot be substituted for the zinc required in the diet. Iron and nickel were similarly ineffective in relieving the zinc deficiency symptoms in rats but did alter the concentrations of zinc in some tissues.[848] Nor did increasing the copper or manganese levels affect symptoms of zinc deficiency.[1194]

Although cadmium normally is neglible in foods,[1102] a few studies have been done of zinc-cadmium interactions in the diet.[1236] Zinc or zinc plus copper[1047] in the diet protected against toxicosis from dietary cadmium. Distribution of injected cadmium was not different in weanling hamsters fed a low-zinc diet compared to those fed a stock ration.[1045a] But the hamsters on the low-zinc diet were apparently not very zinc-deficient. Injecting cadmium on day 12 into pregnant rats caused more resorptions in females fed a low zinc (7 ppm) diet on days 4-12 than in those fed a high zinc (135 ppm) diet.[1211a] Cadmium decreased the intestinal uptake of zinc-65 in rats and calves,[448] but in the deficient rats the transport of zinc to the carcass was not inhibited.[1352] Nor did copper inhibit zinc transport in zinc-deficient rats, although it had some effect on rats with adequate zinc levels.[448] Since more orally administered zinc-65 was excreted by copper-deficient rats than by controls, it has been hypothesized that copper

facilitates zinc absorption.[19] The difference was small, however, and could have been caused by differences in growth or food consumption. There were no differences in zinc uptake and transfer by everted intestinal sacks of copper-deficient rats or the controls.[1456] However, binding of copper and zinc by liver proteins has been shown to be related.[155a,156a,156b]

Form of Zinc

Absorption or retention of zinc was about the same in rats, cattle, and quail whether the intake was in the form of an inorganic salt or incorporated in a natural food.[911,1122a,1128,1478] The anion associated with the zinc does not affect utilization of zinc,[32] although its solubility in the alimentary tract of sheep varies with pH.[155] Zinc sulfate has been reported to result in greater weight gains in pigs than did zinc oxide,[51] but the data are not very convincing since the pigs supplemented with zinc sulfate did not grow any better than those supplemented with magnesium sulfate and no zinc.

OCCURRENCE OF ZINC DEFICIENCY

The extent to which zinc deficiency, particularly marginal zinc deficiency, occurs in man and animals outside the laboratory is largely unknown. The chief symptoms of zinc deficiency--decreased food intake and cessation of growth--are not, of course, unique to zinc deficiency. A test which will distinguish zinc deficiency from other deficiencies and is suitable for screening large populations of animals has yet to be developed. Because of the widespread presence of zinc in foodstuffs, zinc deficiency was long considered not to be a problem of any practical importance.

Increased use of supplements such as soybean meal in animal diets, particularly for pigs and chickens, produced demonstrable zinc deficiencies despite supposedly adequate amounts of zinc in soybeans. Diets for these animals are now routinely supplemented with zinc.[1049] Only a few severe cases of apparent zinc deficiency in cattle have been reported,[1050] but unrecognized cases of mild deficiency may be extant, particularly in areas where the zinc content of forages is low. Crested wheat grass (Agropyron cristatum) in northern Nevada was reported to have a content of 13 ppm zinc,[125] less than that required by calves on a purified diet to maintain normal serum zinc levels. Zinc levels as low as 6 ppm have been found in some grasses (Kubota, unpublished data). Animals grazing on such grasses might well suffer from zinc deficiency.

It is now known that zinc deficiency may occur in humans, particularly in people who derive much of their protein from plant sources. A recent report of apparent zinc deficiency in middle-class American children[617] indicates that the problem is more widespread than had been assumed. Forty-nine percent of children selected for short stature in a group of Project Head Start children had hair low in zinc.[1723] Similarly, about half the girls from low income families in another study had somewhat depressed zinc levels in their hair,[1217] although no correlation with growth seemed to exist.* Hair zinc was low in Iranian[248] and Egyptian [248,1558] hypogonadal patients with retarded growth.

ASSESSING ZINC STATUS IN ANIMALS

Because signs of a marginal zinc deficiency generally are nonspecific, some other test of zinc status is needed. Attempts to correlate zinc

*See Burch et al.[201a]

concentration in blood, hair, or urine with zinc status have met with limited success, as have endeavors to correlate zinc status with enzymes or metabolites in blood or urine. There have been recent attempts to correlate zinc status with taste acuity and salivary zinc levels. The problems involved in using these measures are discussed below.

Plasma Zinc

Although red blood cells contain significantly more zinc than do plasma or serum, zinc in the red blood cell appears to be firmly bound and shows little change in deficiency. Plasma or serum zinc is more variable and does reflect changes in the zinc status of the individual. Serum or plasma zinc in rats, for example, fell significantly after only 1-2 days on a deficient diet,[1199,1773] and it remained low throughout the period of low zinc intake.[199] This pattern was also true in humans. Low plasma zinc levels have been linked to poor growth and symptoms of zinc deficiency in cattle and sheep, although it was pointed out that at least two low serum zinc levels obtained on separate occasions would provide more conclusive evidence than a single determination.[1064] Unfortunately, many other conditions are responsible for reduced plasma zinc levels, including pregnancy, various diseases, and stress.[614] Food intake also affects serum zinc level. The plasma zinc levels of zinc-adequate quail[634] and pigs[880] and zinc-deficient chicks[821] dropped when they were refed protein-containing diets after fasting. An oral protein supplement lowered serum zinc in healthy adults.[1235] Administration of zinc sulfate to fasting subjects increased serum zinc levels more than did administering zinc with a meal.[3,1216a]

Measurement of plasma zinc levels after oral dosing with zinc has been suggested as a better method for determining zinc deficiency than the usual measurement of plasma zinc, as rats on a zinc-deficient diet had higher levels of zinc in plasma after dosing with zinc than did

1774
those that had been on the control diet. The situation is consistent with the observation that everted jejunal sacs (intestinal segments) from zinc-deficient rats took up more zinc than did those of fully fed controls.[850,1456] Other suggestions have been made that zinc absorption is increased in zinc deficiency,[1048] but the evidence is not conclusive. Uptake by animals on restricted intake was also altered, however, suggesting that any animal receiving less than a normal food intake might also show a higher level of plasma zinc after dosing. Thus it is unlikely that determining plasma zinc levels after oral dosing offers any significant advantage.

In summary, fairly severe zinc deficiency would be reflected in a low plasma zinc concentration, but marginal deficiencies are unlikely to be diagnosed by this index. For example, in cases of total starvation, plasma zinc levels were not lowered although body zinc stores were being depleted, indicated by excretion of zinc in the urine.[1532]

Hair Zinc

Because hair samples are easy to collect and store, their use for estimates of zinc status has been looked into by several investigators.[504,614] Various washing procedures were used to remove surface contamination, sweat, and body oil from the hair. One study of such procedures concluded that none of them satisfactorily overcame the effects of the cosmetic treatments commonly used on human hair.[701] With care, however, reliable estimates of zinc content of human hair probably can be obtained,[1525] although it may be wise to exclude samples subjected to the more severe treatments

such as permanent waving, bleaching, and dyeing. Use of pubic hair has been suggested in cases where values from scalp hair might be unreliable because zinc concentrations in scalp and pubic hair of pregnant women have been found to be the same.[89]

It is necessary to discern how useful the zinc content of the hair is in determining zinc status. The zinc content of hair is a reflection of chronic zinc status over the time the hair has been growing, because hair zinc does not exchange with the body zinc pool. Hence it is no surprise that no correlation was found between the zinc concentrations in plasma and hair of Iranian village children.[999] The lack of correlation between plasma and hair zinc was demonstrated more strikingly by an acrodermatitis enteropathica patient whose plasma zinc levels were very low (less than 0.4 μg/ml), but whose hair zinc level was in the normal range. The normal hair zinc level may be related to the decreased hair growth in these patients.[1037a] An extensive study of hair zinc in young rats on varied levels of zinc intake showed that a diet deficient enough to prevent weight gain also prevented hair growth, and therefore zinc concentration did not change substantially.[1198] A level of dietary zinc that was still suboptimal, yet which permitted some growth, resulted in increasing hair growth and reduced zinc concentration in the hair.[1198] Lack of correlation between plasma and hair zinc was also reported in a Panamanian population,[855] although hair zinc in the females varied with geographic location.[856] Zinc concentration in human hair is also reported to vary with age.[617,1235b]

Hair zinc content in female goats was suggested as a good indicator of zinc status because concentrations were decreased in animals on the deficient diet, but the concentration only was measured when the goats were

severely deficient.[569] Zinc concentration was not correlated with the color of bovine hair in samples collected from field locations,[1150] but it has been reported to vary with the season.[393a]

Fairly severe zinc deficiencies in animals, particularly over a long period of time, would probably be reflected in a low hair zinc concentration. Marginal deficiencies are unlikely to be diagnosed in this manner although a study of hair zinc in middle class American children[617] indicated that the zinc level of hair might be a useful measure in detecting deficiency in humans.

Urinary Zinc*

Although zinc is largely excreted in the feces--with only a small amount excreted in urine--some indication exists that, at least in man, urinary zinc may be decreased in states of deficiency. Urinary zinc has not been measured extensively in animals, although urinary zinc excretion was studied in rats after EDTA administration to determine whether this technique might be a way of assessing zinc stores.[896] Such a procedure would be useful primarily for laboratory studies.

Zinc Enzymes

The existence in zinc-deficient animals of an inactive zinc-dependent enzyme that could be activated by the *in vitro* addition of zinc would provide an excellent assay for zinc deficiency. For example, a mutant of the bacterium *Escherichia coli* grown in low zinc media produces an inactive form of the enzyme alkaline phosphatase, which can be activated if the enzyme is incubated with zinc.[602] The existence of this type of enzyme in blood would be obviously advantageous to diagnosis of zinc deficiency. Unfortunately, such an enzyme has not been found in animal tissues.

*See also Chapters 7 and 10.

Zinc-dependent enzymes in blood are of the most interest for their potential in assessing zinc status. Alkaline phosphatase is one of the enzymes that has been measured extensively in young, zinc-deficient animals. The enzyme appears to be consistently reduced in serum of zinc-deficient pigs [11,349,350,1146,1285] compared to controls, but in most cases the controls have grown more than the deficient animals. However, an instance of zinc deficiency in pigs was reported in which serum alkaline phosphatase was not decreased. [201b] The results with rat serum have been more variable [742,849,1370] and not always different from controls on a restricted food intake, suggesting that decreased growth influenced the decreased activity. Since alkaline phosphatase in serum comes primarily from bone and liver, [1765] slight differences in growth rate may significantly affect the amount of the enzyme in serum. However, in a 22-yr-old acrodermatitis enteropathica patient presumably not in an active stage of growth, the level of serum alkaline phosphatase rose parallel to the increase in serum zinc when zinc treatment was applied. Addition of zinc to serum *in vitro* did not increase the activity. [1129a] Alkaline phosphatase was also low in serum of a boy who appeared to be zinc-deficient by other criteria, [615] but not in Iranian boys with low plasma zinc levels. [1363] Calves with hereditary zinc deficiency were observed to have only slight alterations in alkaline phosphatase activity, although serum zinc levels decreased to 0.4 μg/ml. Symptoms of deficiency appeared 2-3 wk after the serum zinc fell to that level. [881a] Since the activity of alkaline phosphatase can be altered by other conditions which interfere with normal metabolism of bone or liver and because low levels are usually found along with low

levels of serum zinc, serum alkaline phosphatase is of no greater diagnostic value in determining possible zinc deficiency than the measurement of serum or plasma zinc level.

Other zinc enzymes, such as lactic and malic dehydrogenase,[1370] have been measured in serum of zinc-deficient animals, but their enzymatic activity was not affected. Carbonic anhydrase activity was lower in deficient animals if the increase in numbers of erythrocytes from hemoconcentration in the deficient animals was taken into account.[1374] Since restricted-feed intake can also cause hemoconcentration, decreased activity was probably related to the reduced food consumption of the deficient animals as was, in fact, found in another study.[742]

Therefore, no presently known enzyme is a reliable measure of zinc status.

Metabolites

Concentrations of various metabolites in blood and/or urine have been reported to be changed in zinc deficiency, but none of the changes appear to be observed consistently in deficient animals and are therefore of little value as a diagnostic tool. Metabolites that have been reported to appear in increased concentrations are uric acid in blood[1783] and urine;[730] hydroxyproline,[729] sulfate,[731] taurine,[733] and alanine in urine;[567] and taurine[567] and free fatty acids in plasma.[1308] Urinary ascorbic acid has been reported as decreased.[93] All these metabolites have been observed in young animals, and it is quite possible that they are nonspecific changes related to the slow rate of growth caused by the zinc deficiency. Reduced

plasma protein in severely zinc-deficient rats was not related to decreased growth or food intake;[1370,1598] however, neither serum protein nor albumin levels were lowered along with the very low serum zinc level of patients with acrodermatitis enteropathica.[1129a] Plasma protein would be affected by many conditions other than zinc deficiency.

Taste Acuity and Salivary Zinc

Because some conditions of abnormal taste acuity have been zinc-responsive, both taste acuity and salivary zinc have been measured in connection with studies of possible zinc deficiency. Taste acuity was impaired in children with poor growth and low levels of hair zinc,[617] but not in a young woman with acrodermatitis enteropathica, a zinc-responsive disease in which plasma zinc levels are very low.[1129a] Zinc secretion in parotid saliva was lower in children who had low levels of hair zinc than in normal children.[615] The data are insufficient at present to indicate whether either of these measures is a reliable and specific indicator of zinc status. In any event, measurement of taste acuity would not be a practical means of screening large populations. Salivary zinc could be measured on animals as well as people. The large variation in zinc concentration of saliva collected from the same individual at different times[1536] suggests that there may be substantial problems in the use of this criterion. Nonetheless, in view of the striking changes that have been reported in the tongue of zinc-deficient sheep[973] and monkeys,[79] in the mouth[1180] and esophagus of pigs,[1146] and in zinc-deficient rats[461,1102a] (including the fetal rat),[382] measurement of salivary zinc may be worth pursuing.

METABOLIC CONSEQUENCES OF ZINC DEFICIENCY

Although many studies have examined in some detail the effects of zinc deficiency on various systems in the animal, the critical roles of zinc in these systems still are not known. Zinc is contained in a number of enzymes, but the effect of zinc deficiency on the activity of these enzymes is variable. Also, decreased activity of the known zinc-dependent enzymes seems not to be a major factor in the disturbances observed in zinc deficiency. Because some of the earliest effects of zinc deficiency are reduced food intake and cessation of growth, it is very difficult to distinguish between a primary effect of a lack of zinc on the animal and secondary effects based on decreased food consumption and halted growth.

In theory, it is possible to control for the decreased food consumption by feeding a zinc-adequate animal the same amount of food that the zinc-deficient animal eats. The problem, however, is not so simple as it might appear, since the deficient animal has a cyclical pattern of eating,[270,271,1782] will gain weight less quickly than a zinc-adequate animal on the same amount of food, and will consume its food over a 24-h period, whereas the pair-fed animal consumes its food in a much shorter time. To control for all these effects is difficult and often is not done. Since food intake can influence metabolism in ways and over time periods that are not obvious, differences between deficient animals and their "controls" must be interpreted very cautiously.

DNA, RNA, AND PROTEIN SYNTHESIS

Because of the rapid effect of a lack of zinc on growth, a number of studies have been conducted on DNA, RNA, and protein synthesis during

zinc deficiency. Concentrations of DNA, RNA, and protein were not remarkably different in the deficient animal,[467,493,494,775,878,1172,1280,1284,1285] but the concentration of these compounds may be less important than their metabolic activity. The activity of these compounds has been estimated by measuring the incorporation of radioactive precursors into DNA, RNA, or protein. In order for such measurements to be reliable, however, the radioactive precursor must have the same relative concentration in both deficient and adequate animals. Since concentrations of free amino acids were higher in both skin[732] and plasma[567] of deficient animals than in controls, and since compounds such as thymidine[729a] and several amino acids[733a,1607a] were metabolized to carbon dioxide to a greater extent, the radioactive precursor may constitute a smaller proportion of the precursor pool in the deficient animal than it does in the control. Different concentrations of the radioactive precursor may be brought about by differences in enzymes needed to enable the radioactive compound to enter the precursor pool, as well as by differences in the concentration of metabolites in the cell. For example, incorporation of radioactive thymidine into the pool of DNA precursors in the cell depends on the activity of thymidine kinase, an enzyme whose activity has been shown to vary diurnally,[722] which is related somewhat to food consumption.[351a] The different eating pattern of the zinc-deficient animal therefore could make a difference in the time at which thymidine kinase activity occurs and consequently in the time at which peak incorporation of thymidine would be observed. In rats on a low zinc diet, the peak of thymidine incorporation which follows partial hepatectomy was delayed compared to the controls, but there was little difference in the amount of incorporation at the

respective peaks.[397,408] Others have reported differences in thymidine incorporation between zinc-deficient animals and controls in which the magnitude of the differences changed with time.[732,1002,1550] Thymidine kinase activity, as measured by thymidine incorporation into DNA *in vitro*, was reduced in connective tissue from animals on a low zinc diet; the concentration of zinc used in the assay systems differed, however, and the animals on the low zinc diet also were receiving 2.5 times as much phytate as the controls.[1280]

DNA

Thymidine incorporation was reduced in liver, kidney, and spleen in young rats before growth and food consumption were affected by the low zinc diet.[1779] The sensitivity of these tissues to the early effects of the deficiency was no doubt related to the rapid turnover of zinc in these tissues.[920] Thymidine incorporation was also reduced in 12-day-old embryos of female rats given a deficient diet. However, mitoses in neuroepithelium were increased.[1584] In rapidly differentiating tissue such as embryonic tissue, tissues from animals fed a deficient diet may not be in the same stage of development as the controls although the time of gestation is the same. Different stages of development at the same time of gestation were observed between fetuses from folic acid-deficient females and fetuses from controls.[788a]

Thymidine incorporation was decreased in zinc-deficient rats more than in controls after wounding the skin[736] or implanting sponges under it,[1280] but not after an esophageal wound.[966] Interpretation of studies of incorporation after surgery is complicated by the hormonal changes which accompany stress, since adrenal hormones, for example, affect thymidine incorporation.[542a]

In contrast to reduced mitotic activity observed in most tissues of the zinc-deficient animal,[383,461,1180] mitotic activity in the esophagus and buccal mucosa[23] was stimulated. The stimulation may be peculiar to zinc deficiency because it did not occur in pair-fed controls.[383,461] Furthermore, a single dose of zinc in zinc-deficient rats reduced cell division in the esophagus but increased the mitotic indices of epidermis and liver.[461] In addition to increased mitotic activity,[23] thymidine incorporation,[24] and dry weight[1037] of cells in buccal mucosa of deficient rats, epithelial thickness was increased.[1180] The increase in epithelial thickness occurred before the increase in mitotic activity and was thought to come from interference with the normal shedding mechanism, perhaps from an increased glycoprotein coating of keratinizing cells.[23] Apparent interference with the shedding mechanism was also observed in esophagi from 18-day-old rats nursed by zinc-deficient dams[1102a] and in pigs on a low zinc-high calcium diet.[1146] Alterations in salivary and intestinal mucus have been reported in zinc-deficient rats.[1309]

Cell cultures also are affected by a lack of zinc. Results in these systems are clearer, because differences from food intake and hormonal changes are eliminated. Addition of a zinc chelating agent inhibited incorporation of thymidine into DNA in primary rabbit kidney cells,[931a,931b] in human[1785] and porcine lymphocytes,[269] and in chick embryo cells.[1377,1378,1379] However, the zinc requirement was lost rapidly by rabbit cells, and continuously cultivated cells such as HeLa cells and L cells were also resistant to EDTA.[931a] Chick embryo cells retained the requirement for zinc but became quite resistant to EDTA after infection with Rous sarcoma virus.[1377]

The effect of EDTA did not appear to be caused by damage to the cell membrane since cell movement and uptake of glucose were normal.[1379]

Although good evidence exists that both DNA-[1494,1542] and RNA-dependent DNA polymerases from some organisms are zinc-dependent[59,59a,59b,1255] enzymes, EDTA probably does not affect enzymes directly involved in DNA synthesis. A delay in the addition of EDTA decreased its effect in several systems,[268,269,515,931a,931b] and in continuously replicating cells EDTA was able to reduce thymidine incorporation only after a time lag.[1377] Both these results are interpreted to mean that zinc is required for a critical step preceding the onset of DNA synthesis in the cell, and that cells past this step when EDTA is added will replicate normally.[268,931b,1377]

Studies of microorganisms grown in low-zinc media are also consistent with this hypothesis. Although growth of Mycobacterium smegmatis was inhibited by a low-zinc medium, the activity of DNA polymerase in vitro was not affected.[201a] As has been pointed out, an enzyme which is zinc-dependent in one species need not be so in another; however, the activity of the polymerase from Mycobacterium smegmatis was inhibited by a zinc chelating agent and was presumably a zinc-requiring enzyme.[1790] Euglena gracilis also appeared to synthesize DNA normally, but it failed to divide.[452] The DNA synthesized could, of course, have been abnormal. In the zinc-deficient Mycobacterium smegmatis, increased DNAase activity was suggested to be related to increased need for DNA-repairing enzymes to compensate for synthesis of abnormal DNA. DNA synthesis was not affected in Rhodotorula gracilis.[292]

In summary, the increased mitotic activity in esophagus, buccal mucosa, wounded skin, and liver after partial hepatectomy, as well as the

synthesis of connective tissue in response to an irritant, provide ample evidence that zinc-deficient animals can synthesize DNA. The lack of growth in zinc-deficient animals seems to be due to an effect on factors initiating DNA synthesis rather than to any defect in the process once it has begun.

RNA

In general, incorporation of precursors into RNA is less affected by a lack of zinc than is incorporation of precursors into DNA. Levels of EDTA that almost completely inhibited thymidine incorporation into DNA had little effect on incorporation of precursors into RNA in rabbit kidney cells,[931a,931b] human,[1785] or porcine[268,269] lymphocytes, and perfused liver.[515] EDTA reduced the incorporation of precursors into RNA in chick embryo cells to 60% of normal, whereas incorporation of thymidine into DNA was almost completely inhibited.[1377] Incorporation of uridine into brains of both zinc-deficient rats and controls was identical.[1172] Synthesis of RNA was not affected in Mycobacterium smegmatis grown in a medium low in zinc,[635a] but it was decreased in Rhodotorula gracilis[292] and Euglena gracilis.[452]

RNA polymerase has been reported to contain zinc,[1462] but its activity was less inhibited by EDTA the later the EDTA was added to the incubation mixture. This phenomenan has been interpreted to mean that zinc is required for the initiation of RNA chains but not for their elongation. Because the enzyme does contain zinc, its activity has been measured in zinc-deficient animals. Its activity was reduced in liver[1603] and brain[493] from rat pups nursed by zinc-deficient dams. However, the pups continued to grow despite the reduced polymerase in the liver;[1603] and the activity in brain, although reduced at 6 days, was no longer reduced at 16 days.[493] RNA polymerase activity in rat liver nuclei was also affected by starvation,[122a,1345b]

but the relation between RNA synthesis in the animal and the measurement of RNA polymerase in the test tube is not clear. Zinc stimulated orotate incorporation *in vivo* without any detectable effect on RNA polymerase activity measured *in vitro*.[1757] Zinc inhibited polymerase activity *in vitro*.[1757,1757a] The increase in material in the monosome region of the sedimentation profiles of zinc-deficient rat pups,[493] starved pigeons,[122b] and mice[1144a] could represent defective RNA synthesis, but it may simply reflect the decreased protein synthesis that is apt to follow decreased growth. Neither additional polymerase[1345b] nor initiation factors[122b] increased RNA synthesis in preparations from starved animals, further suggesting that reduced polymerase activity was the result rather than the cause of decreased growth.

RNAase activity was increased in testes of rats that had been on a deficient diet for 5[1284] or 11[1522a] wk. Since the testes of growing rats are particularly susceptible to zinc deficiency, increased RNAase activity at these relatively late stages of the deficiency may also have been a result rather than a cause of the problem. RNAase activity was not different in brains of rats nursed by females on a low zinc diet, although brain growth itself was reduced.[494]

Protein Synthesis

Nor did EDTA have much effect on incorporation of labeled amino acids into protein in rabbit kidney cells,[931a] chick embryo cells,[1377] and perfused rat liver.[515] Since EDTA only affected amino acid incorporation into protein in lymphocytes at later stages of the cell cycle (but not initially),[269,1785] EDTA presumably did not affect protein synthesis *per se*. A zinc-deficient diet also had little effect on incorporation of labeled amino acids into protein in young rats.[408,1172] Incorporation of several amino acids,[533,731,735]

including selenium analogs,[1003] was decreased in skin from deficient rats but not in other tissues. Incorporation of radioactive amino acids into liver, heart, and kidney was reduced in rat pups nursed by females on a low zinc diet.[496] Aside from the differences in incorporation in skin and in nursing pups (perhaps partly caused by differences in amino acid metabolism[533] by the deficient animals), little indication has been found that a zinc-deficient diet interferes with protein synthesis. The deficient rat was able to synthesize new tissue in response to an irritant; and although the total amount synthesized was less than that of controls, the amount synthesized relative to weight gain was as high in the deficient animal as in the ad libitum control.[467,1284] Deficient rats also synthesized protein in response to infection[1218] and after partial hepatectomy.[267a]

INTERMEDIARY METABOLISM

Glucose

Because the concentration of zinc in the pancreas decreases markedly when zinc is deficient, and because the addition of zinc to insulin preparations prolongs their action, several studies have been made on the effect of zinc deficiency on glucose tolerance. Results have conflicted, somewhat accounted for in that glucose tolerance in rats appears to be closely related to the food consumption of the animals in the 24 h preceding the pretrial fast.[479] In studies where differences in glucose tolerance existed,[139,740] comparisons were apparently made to ad libitum-fed controls.[139,1309a] Differences in glucose levels in fasted animals,[663,1221,1308] glucose tolerance,[139,740] insulin levels in blood before and during glucose stimulation,[139,1221, 1308,1309a] and insulin levels in the pancreas[740,1221]

between deficient animals and pair-fed controls generally have been small. Blood glucose was higher in deficient animals that were given a second dose of glucose within 2 h after the first dose.[663,1309a] Pancreatic tissue from deficient animals released less insulin *in vitro* in response to glucose stimulation than did tissue from *ad libitum*-fed controls but the effect was obtained only at a high glucose level.[740] Insulin secretion in response to glucose stimulation is also affected by starvation.[659a] Resistance to insulin coma was greater in zinc-deficient rats,[1309a] but resistance to insulin coma is also affected by starvation.[1594a]

Fatty Acid

High levels of free fatty acid were observed in fasted rats during the first 3-4 wk after they were placed on a zinc-deficient diet.[1308] However, values approached those of the control group with time. Zinc stimulated the glucose uptake by rat adipose tissue *in vitro*,[1307b] although the magnitude of the effect varied with the species of rat.[1307c] Glucose uptake by adipose tissue from zinc-deficient rats was less than that from rats given a zinc supplement.[1307b]

Amino Acid

Amino acid metabolism, particularly that of the sulfur-containing amino acids, has been reported to be disturbed in zinc deficiency. Urinary excretion of both total sulfur and sulfate from sulfur-labeled methionine,[735] cystine,[733] and taurine,[42] as well as sodium sulfate[733] was increased in deficient rats.

The urinary excretion of taurine increased in one investigation[42] but not in another.[567] Hydroxyproline excretion also increased in young zinc-deficient

rats.[729] Levels of free amino acids in rat skin were increased in deficient animals.[732] Activities of the amino acid-catabolizing enzymes arginase and tryptophan pyrrolase were also increased, although the activities of serine and threonine dehydratases were not affected.[730] In these experiments, the weight of the pair-fed animals exceeded that of the zinc-deficient ones. Therefore, the differences observed may have been due to differences in growth rate of the two groups of animals. Both food restriction and zinc deficiency affect zinc-binding proteins in rat liver.[156,364,1342]

ZINC DEFICIENCY AND BONE FORMATION

Bone abnormalities in zinc-deficient chicks and rat embryos have led to studies on the effects of zinc deficiency on calcium metabolism and bone growth.[99,100,102,221,755,949,951,1139,1146,1760] Kinetic studies of calcium-45 in zinc-deficient rats showed that turnover of calcium was slower in deficient than in pair-fed animals and that pool size was smaller. Absorption and excretion were not different, however.[755] Sulfate uptake and turnover in growing regions of bone were impaired in the zinc-deficient chick.[1139] *In vitro* sulfate uptake by skin from pigs given a moderately low zinc diet was not impaired.[1610] There was no difference in collagen or mucopolysaccharides of zinc-deficient chicks, although the hexosamine content of primary spongiosa was somewhat higher.[1139] Hexosamine content in bone from deficient rats was not different from the controls,[949] although hexosamines in saliva were affected.[1309] No change occurred in the hexosamine content of skin from pigs given a moderately low zinc diet.[1610] Many changes that were observed in bone from deficient animals[99,102,949,1146] were similar to those observed

with decreased feed intake. Zinc and manganese appeared to interact in otolith formation in mice.[1306a]

Zinc deficiency in chicks causes a swollen hock condition which resembles arthritis. Certain anti-arthritic compounds relieved the condition[715] while others did not.[1323] How the effective antidotes acted was unclear, although in some cases sulfate uptake in areas of bone growth increased. However, sulfate uptake increased in adequate as well as deficient chicks.[395]

Increased dietary histidine also relieved the swollen hock condition and was postulated to function by increasing histamine concentration, but histamine in tissues of deficient chicks was not different from controls, nor was it increased by the feeding of histidine.[1324] Histidine did increase alkaline phosphatase activity measured in the entire tibia,[904,905] although histologic studies did not show any difference in the distribution of alkaline phosphatase in the epiphyseal plate.[1760] Some changes in bone histology were apparent before alkaline phosphatase would be produced,[1760] so that deranged alkaline phosphatase may be a symptom rather than a cause of problems in bone formation.

ZINC DEFICIENCY AND ZINC-RELATED ENZYMES

As with the DNA polymerases, the demonstration of a zinc-requirement by an enzyme in an organism has led to its measurement in zinc-deficient animals, although the enzyme in the animal may not have been shown to be zinc-dependent. Again, it must be emphasized that the metal requirement for the same enzyme in different species need not be the same. A comparison of

activities of various enzymes in zinc-deficient animals in relation to the controls is given in Tables 8-4 and 8-5. The tables only are intended to indicate whether a difference was observed between the activity of the enzymes in the tissues of zinc-deficient animals and those of controls; and, if values were given for both pair-fed and ad libitum controls, whether the enzymatic activity in the tissues from the pair-fed animal varied in the same direction as that of the deficient one. If a change is shown in the deficient animal without a notation of change in the pair-fed control, it may mean that information on pair-fed controls was not given. Also, increased enzymatic activity in the pair-fed control relative to the activity in the ad libitum will not be reflected in the tables unless the enzyme activity was increased in the deficient animal.

It should also be pointed out that different authors expressed their results differently. Ratings in the table were based on activity/unit weight rather than on total activity, since the total would obviously be influenced by the smaller size of the tissues in the deficient animals. Nonetheless, activity/unit DNA was significantly different in some tissues in one study,[1285] whereas activity/unit protein was not. Also, in one of the studies, pair-fed controls were not used because in a previous study they had become "zinc-deficient."[775] It seems more likely that the animals were biotin-deficient, as diets containing egg white increase the requirement for biotin. Since egg white protein is widely used in studies of zinc deficiency because of its low zinc content, increased biotin requirement should be taken into consideration although most investigators do add an excess of biotin to such diets.

As shown in Table 8-4, the activities of presumably zinc-related enzymes in the deficient animal have varied from tissue to tissue and from experiment to experiment. In Table 8-5 it can be seen that other enzymes not known as zinc-dependent enzymes were also altered in the zinc-deficient animal.

Alkaline phosphatase in bone was consistently lowered in the zinc-deficient animal, as was alcohol dehydrogenase in liver and carboxypeptidase in the pancreas. The volume of pancreatic secretion was also smaller in zinc-deficient pigs.[1565] The decrease in α-mannosidase in serum may be useful as an indicator of zinc status because the activity of the enzyme in liver was reported to decrease in zinc-deficient rats and be activated by zinc *in vitro*, whereas the activity of the liver enzyme from zinc-adequate rats was only slightly stimulated.[1213a] As mentioned, this type of effect would make a useful assay of zinc deficiency.

Little correlation has been found between zinc concentration in tissues and the activity of the presumed zinc-dependent enzymes. Concentration of zinc in the liver was not lowered in the deficient animal in two of the studies,[1279,1372] whereas liver alcohol dehydrogenase was lowered. The activity of alcohol dehydrogenase correlated well with the concentration of zinc if food were restricted but not if animals were fed normally.[1330]

Because enzymatic activity is not correlated with zinc in the tissues and most enzymes are not activated by zinc in *in vitro* assays, it is possible that enzymatic activity is low in the zinc-deficient animal because the animal has stopped growing and consequently needs less enzyme. In any

event, of the few enzyme activities that have been consistently lowered, none seem likely to account for the symptoms (see Table 8-1) observed in the deficient animal.

Alkaline phosphatase has been one of the most extensively studied enzymes in zinc-deficient animals. Determination of alkaline phosphatase activity in animals is complicated by the fact that enzymes from different tissues varied in pH optima, activity with different substrates,[783a] and activity in presence of magnesium.[361a,361b,783a,948a] Within a single tissue several isoenzymes may exist; as many as five have been reported in intestinal mucosa.[361a] Only one isoenzyme--with a pH optimum at 10.5--was activated by adding zinc *in vitro*,[783a,1326a] although intestinal homogenates have not been activated by *in vitro* addition of zinc.[1778] In cases where alkaline phosphatase was activated, it happened to the control as well as the deficient enzyme. Chromatography of homogenates from deficient and control animals indicated that the same isoenzymes were present in both animals, although some properties of the enzyme from the deficient animal differed from those found in the control.[1326a] The apparent differences, however, may have been a consequence of differences in degree of purification of the preparations.

ZINC AND REPRODUCTION

Another major effect of zinc deficiency is its interference with the growth or function of the reproductive organs. This is another area in which it is extremely difficult to distinguish between the effect of a lack of zinc *per se* and the effect of reduced food intake.[907a] Underfeeding is well known to interfere with reproductive function.

Female

In the rat, inadequate zinc severely affects reproduction.[220,327,759,761,1582,1640,1732] Effects include difficult parturition, congenitally malformed young, failure to maintain pregnancy, and cessation of estrous cycles. Congenital malformation or difficult parturition occurred after consuming a low zinc diet[46,756,1731] or large amounts of a zinc chelate[1582] for just a few days during a critical period of gestation. Development of preimplantation eggs was abnormal in females fed a deficient diet for only the first few days of pregnancy;[758] however, offspring born after dams received a deficient diet for days zero to five had only a slight incidence of malformations.[756] Work with the teratogenic effects of trypan blue also suggest that there is some mechanism for repairing early damage to the eggs.[621b]

Administration of zinc late in pregnancy prevented stress at parturition,[47] but survival of offspring from dams fed a zinc-deficient diet during the second week of gestation was poor, although zinc nutriture after that was adequate.[757] Survival of pigs born to gilts on a low zinc diet for approximately the last third of pregnancy was also poor,[1744a] and abnormalities in bone development were observed in the fetuses.[1202] Feeding the rat a zinc-deficient diet during lactation caused severe zinc deficiency in the pups.[1102a]

Insufficient zinc during pregnancy also interferes with reproduction in the rabbit.[44] Studies of zinc metabolism in zinc-adequate rabbits have shown that zinc accumulation in the rabbit endometrium coincided with the blastocyst phase of embryonic development.[958a] About that time, luteal tissue also showed a marked increase in specific activity of injected zinc-65,

although the zinc concentration in the tissue did not change.[1011a] Turnover of zinc-65 in the endometrium decreased in the pregnant, pseudopregnant, and superovulated rabbit. In placental tissues, zinc transport varied with gestational age, and fetal placenta exchanged zinc with blood plasma four times faster than maternal placenta.[1011] Fetal plasma in rats,[757] pigs,[1202] and sheep and goats[929a] is higher in zinc concentration than maternal plasma. The concentration of zinc in human amniotic fluid at the end of pregnancy has been correlated with the birth weight of the newborn.[453a]

Zinc *in vitro* has been shown to potentiate contractile responses of rat uteri to submaximal doses of acetylcholine.[354b] Since uterine contractions have not been measured in the zinc-deficient pregnant animal, it is not known whether there is anything abnormal about the contractions that could be related to the difficulty at parturition. Zinc concentration in the deficient uterus was the same as in the controls.[46] But such a condition does not indicate of itself that contractility was normal since the potentiation *in vitro* did not correlate with the zinc content of the uterus.[354b] Adding zinc to an endometrial homogenate has been reported to increase binding of β-estradiol to protein.[435a]

Most studies dealing with the relation of zinc to reproduction in the female have been done with animals that bear large litters, such as the rat, rabbit, and pig. Whether the work with these animals applies to other species, particularly those that bear single offspring during relatively long gestation times, is unknown. For animals that do not have large litters, zinc may be more important for normal mating and maintenance of pregnancy than for normal delivery.

After consuming a low zinc diet for 2-3 wk before mating, rats failed to maintain pregnancy.[45] Consumption of a low zinc diet for longer periods brought about cessation of estrous cycling.[45,544] The effects on the estrous cycle in particular are probably caused by reduced food consumption, although measurements of reproductive hormones indicated that effects of zinc deficiency and restricted food intake were not identical.[544]

Oral contraceptives lower plasma zinc levels in humans[614] and rats.[1000] The percentage of zinc-65 in other tissues of rats given high levels of estrogen was increased somewhat; however, the dose administered signficantly depressed weight gains in the growing females.

Male

The relation of zinc to the male reproductive function has received considerable attention because of the high concentration of zinc present in the prostate gland and semen.[1640] The major portion of the zinc in semen comes from the prostate gland, but its function is not known. Zinc content of the epididymis is also high[1376b] and may function in retarding oxidation of sulfhydryl groups in the sperm.[224] But sperm motility is not affected by large differences in zinc content,[789a] although sulfhydryl groups in sperm flagellum have been thought to be involved in motility.[224] Zinc also has been postulated to protect the integrity of sperm since oxygen consumption by sperm *in vitro* was raised in the absence of zinc.[427a] Increased oxygen consumption by sperm is associated with metabolic disorganization. Zinc uptake by sperm was affected by factors other than the concentration of zinc in semen.[936a,936b,936c] Interference with normal zinc metabolism was postulated to be a mechanism of the antifertility agent α-chlorohydrin since it increased radioactive zinc in regions of the

testes in which zinc concentration in sperm is normally reduced.[575a] Reproductive hormones have also been shown to alter the distribution of zinc in male reproductive organs.[1640]

Insufficient zinc in growing male rats severely affected testicular development. The effect was apparently independent of the low food intake since pair-weight rats had normal size testes whereas those of deficient rats were considerably smaller than normal.[383] Histologic studies of a few zinc-deficient rats suggested that the earliest effect of zinc deficiency on the testes was the inhibition of the transformation of round spermatids into elongated ones.[1174] Refeeding the animals with zinc for 15 days subsequent to the 28-day depletion period restored the histologic appearance to normal.[383]

However, low food intake does affect the testes in some ways. Alpha-mannosidase activity, for example, has been reported to decrease in epididymal tissue of zinc-deficient rats. But enzymatic activity was restored in castrated animals by injecting them with testosterone, yet zinc concentration was not affected.[1517a] The enzyme may have been affected by the decreased testosterone levels associated with low feed intake.

Libido of adult male goats on a zinc-deficient diet has been reported to be reduced, but the goats may not have had a simple zinc deficiency.[1127]

EFFECT OF ZINC ON FOOD INTAKE

For both animals and man, one of the first effects of a low zinc diet is decreased food consumption.[1004] In rats, food consumption also became increasingly variable and cyclic.[270,1782] Intake varied less if the rats were given a 5% rather than a 20% protein diet. Moreover, zinc-depleted rats ate more of a zinc-supplemented diet only when it contained protein.[270] Examination of the amino acid pattern

in plasma and urine of animals fed low zinc diets containing either 24% or 6% protein indicated that plasma amino acids--particularly nonessential ones--were elevated on the low zinc diets. Tyrosine was the only amino acid that fluctuated significantly in relation to daily food intake.[567] Animals on a low protein diet may not be functionally zinc-deficient, because growth is restricted by the lack of protein and the increased catabolism of body protein may release enough zinc to satisfy temporary maintenance requirements. Mean plasma zinc concentrations of rats on a low protein diet were appreciably higher than in those fed 20% protein diets.[1376a]

Zinc-deficient rats drank more of an acetic acid solution if they previously had received zinc in the solution.[276] Zinc-deficient rats also drank more sodium chloride, hydrochloric acid, and quinine sulfate solutions than did pair-fed controls, and took in a greater total volume of fluid, including tastant solutions and water.[1004] Zinc-depleted chicks also chose a zinc-supplemented diet in preference to a low zinc diet;[746a] after 5 days, however, the controls also selected the supplemented diet. Turkey poults have been reported to select a zinc-supplemented diet,[1699a] but apparently because of a position preference. Food consumption rose within a few hours after zinc repletion in young zinc-deficient rats,[1195] but it increased more slowly in zinc-deficient pregnant rats who were transferred to a zinc-adequate diet.[601]

EFFECTS OF ZINC DEFICIENCY ON WOUND HEALING AND TUMOR GROWTH IN ANIMALS

Reports of beneficial effects of zinc on wound healing in man have led to studies of wound healing in both zinc-deficient and zinc-adequate animals. Wound healing was impaired in zinc-deficient animals;[736,1154,1408] differences were more apt to occur during later stages of healing.[1264] The level of zinc in wound fluid and wound plasma has been reported to increase

in rats [930] and dogs [931] in early stages after wounding. However, the increases observed were highly variable, as were the effects of different stresses on serum zinc in dairy cows.[1745] Additional zinc generally has no effect [427,571,1307,1408] or only slight effect [902,1034,1082] on wound healing in zinc-adequate animals. Zinc sulfate did not improve collagen biosynthesis or fibroblast proliferation in human tissue culture.[1737b] Zinc seemed to have some protective effect on the livers of rats injected with carbon tetrachloride[427] and on the thyroid glands of pigs given methylthiouracil, a thyroid antagonist.[685] The effectiveness of zinc in stimulating lymphocyte transformation [98,1243] and increasing thymus weight [1307a] suggests that excess zinc might stimulate the immune system; but except for the effects mentioned, beneficial effects of zinc treatments on zinc-adequate animals have not been demonstrated.

Zinc deficiency has been reported to decrease the growth rate of several tumors and increase the survival times of animals inoculated with them. If Walker 256 carcinosarcoma (a rapidly dividing solid tumor) were injected intramuscularly in young male rats that had been on a zinc-deficient diet for 8 days, the number of takes was reduced; survival time was increased and tumor size reduced in those animals in which tumors did grow [381] compared to either pair-fed [1018] or paired-weight controls.[381] Growth of a variant of the Walker 256 carcinosarcoma was reduced in young rats that had been on a low zinc diet since weaning.[1235a] Similar results occurred in mice with the slower growing Lewis lung carcinoma.[380] Survival times after injection with various leukemia tumors were not as markedly affected by zinc deficiency as sarcoma tumors [81,380] and attempts to increase the effectiveness of the zinc deficiency in treating leukemia tumors were unsuccessful.[81]

It is important to realize that in the cases in which tumor growth was reduced in zinc deficiency, the animals were already deficient when the tumor tissue was injected. Transferring animals to a low zinc diet after a tumor is established has not been shown to reduce tumor growth.

Because of the hyperkeratosis and parakeratosis of the esophagus that occur in zinc-deficient animals, a relationship between esophageal cancer and zinc deficiency had been postulated. A marginal intake (7 ppm) of zinc did not, however, affect the incidence of nitrosamine-induced esophageal carcinogenesis in rats.[1683]

ZINC DEFICIENCY AND BEHAVIOR

Zinc-deficient rats have performed poorly on different behavioral tests.[220,601,950,1406] Exploratory activity was decreased and performance impaired in rats given a deficient diet for 7 wk after weaning.[219] However, the rats were apparently ill at the time of testing. Behavior was also affected in offspring of rats given a deficient diet during gestation or lactation although the offspring themselves were given a zinc-adequate diet. Offspring from two rats given a deficient diet during lactation made more errors on an elevated maze than did offspring from two pair-fed rats.[950] Offspring from successive litters of females fed a deficient diet throughout gestation and lactation were less active in an open field test than were controls. The number of deficient rats tested, however, was very small and represented few litters, since not many offspring from the deficient females survived to weaning.[220]

Male offspring of 5 dams on a deficient diet only during days 15-20 of gestation avoided shock less successfully than did offspring from 5 pair-fed

rats. Offspring from both deficient and pair-fed animals, however, were less active and extinguished the response faster than did offspring from ad libitum females.[601,1406] Since female offspring of zinc-deficient dams tolerated shock less well than did those from pair-fed animals,[600] poorer performance of the males in the shock avoidance test also may have been due to a reduced tolerance for shock. Females from pair-fed dams were less resistant to shock than were those from ad libitum females.

These studies suggest that consumption of a zinc-deficient diet during pregnancy or lactation has a residual effect on the offspring. Such an effect has also been attributed to protein deficiency during gestation,[737] but the number of pregnant females used in the zinc studies was small. In addition, the effect of zinc may have resulted from differences in the care given the pup by the dam,[46] the suckling ability of the pup and hence its ability to stimulate lactation,[1816a] or the in utero nutrition of the fetus, although the dams received the same amount of food.

Whatever the source of the difference, one must be very cautious in extrapolating these results to other species since the brain of the rat is relatively immature at birth and may be more susceptible to nutritional insults. Brain maturation, as measured by the activity of the myelin-associated enzyme, 2', 3'-cyclic nucleotide 3'-phosphohydrolase (CNP), was not different for rats suckling females on a zinc-deficient diet.[1303] Studies of DNA, RNA, and protein in brains of animals whose mothers were subjected to gestational or lactational zinc deficiencies have revealed relatively little difference in the concentration of these compounds.[493,494]

TABLE 8-4

Effects of Zinc Deficiency on Tissue Levels of Zinc-Related Enzymes

Enzyme	Blood	Bone	Muscle	Lung	Heart	Thymus	Spleen
Alcohol dehydrogenase[1]		Decrease[a](pig) Decrease[e]	No change[d]				
Aldolase		No change[a](pig) Decrease*[e]				No change[e]	
Alkaline phosphatase	Decrease[g](pig) Decrease[l](pig) Decrease[a](pig) No change[b](pig) Decrease[n](pig) Decrease[p](pig) No change[i] Decrease[s]	Decrease[h](chick) Decrease[k](chick) Decrease[j](chick) Decrease[g](pig) Decrease[a](pig) Decrease[i] Decrease*[r] Decrease[e]	No change[i]	No change[i]	Decrease*[i]	Decrease*[e]	No change[g](pig)
Carbonic anhydrase	Decrease*[i] Decrease[t]	No change[i]	No change[i]	No change[i]	No change[i]		
Carboxypeptidase							
Glutamic dehydrase	No change[i]				No change[i]		
Lactic dehydrogenase	Increase*[i] No change[b](pig) No change[s]	Decrease[a](pig) No change[e]	Decrease*[i] No change[d]	No change[i]	Decrease*[i]	No change[e]	
Malic dehydrogenase	No change[s]		Decrease[d]				
α-mannosidase	Decrease[w]			Decrease[w]	No change[w]		Decrease[w]

TABLE 8-4 (Cont'd.)

Enzyme	Tissues						
	Liver	Pancreas	Stomach	Intestine	Kidney	Testes	Brain
Alcohol dehydrogenase[l]	Decrease[o](pig) Decrease[e] Decrease[f]				Decrease[o](pig) Decrease[e]	Decrease[e]	
Aldolase	Decrease[o](pig) No change[e]				Decrease[o](pig) No change[e]	No change[e]	
Alkaline phosphatase	Increase[j](chick) Decrease[k](chick) Decrease[g](pig) No change[i]	Decrease[e] Decrease[n,i]	Decrease[n,t] Decrease[m]	Decrease[j](chick) No change[k](chick) No change[g](pig) Decrease[i] Decrease[n,o] Decrease[m] Decrease[q]	Decrease[o](pig) No change[g](pig) Decrease[e] Decrease[i] Decrease[m]		No change[k](chick)
Carbonic anhydrase	No change[i]	No change[i]	Decrease[i] Decrease[m]	No change[i] Decrease[m]	No change[i] Decrease[m]		No change[i]
Carboxypeptidase		Decrease[o](pig) Decrease[e] Decrease[x]					
Glutamic dehydrase	No change[i] No change[b](pig) Increase[n,f]			No change[i]			No change[i] Decrease[u]
Lactic dehydrogenase	Decrease[n,i] No change[o](pig) Decrease[n,f]	No change[i] No change[o](pig)	No change[i]	Decrease[n,i]	Decrease[n,i] No change[e]	No change[v] No change[e]	
Malic dehydrogenase	No change[f]					No change[v]	
α-mannosidase	Decrease[w]			Decrease[w]	Decrease[w]		No change[w]

TABLE 8-4 (Cont'd.)

Conditions:

Decrease - Deficient lower than either ad libitum or restricted-fed controls.

Decrease* - Restricted-fed as well as deficient lower than ad libitum controls; difference not necessarily significant.

No change - Deficient not different from ad libitum controls.

Increase - Deficient greater than ad libitum controls.

Increase* - Restricted-fed as well as deficient greater than ad libitum controls; difference not necessarily significant.

[a]Studies done in the rat unless otherwise noted.

[b]Derived from Burch et al.[201b]

[c]Derived from Prasad et al.[1285]

[d]Derived from Roth and Kirchgessner.[1376]

[e]Derived from Prasad and Oberleas.[1279]

[f]Derived from Roth and Kirchgessner.[1372]

[g]Derived from Agergaard and Palludan.[11]

[h]Derived from Lease.[904]

[i]Derived from Huber and Gershoff.[742]

[j]Derived from Davies and Motzok.[361b]

[k]Derived from Lease.[905]

[l]Derived from Norrdin et al.[1146]

[m]Derived from Iqbal.[775]

[n]Derived from Dahmer et al.[349]

[o]Derived from Iqbal.[783a]

[p]Derived from Dahmer et al.[350]

[q]Derived from Williams.[1778]

[r]Derived from Roth and Kirchgessner.[1373]

[s]Derived from Roth and Kirchgessner.[1370]

[t]Derived from Roth and Kirchgessner.[1374]

[u]Derived from Prohaska et al.[1303]

[v]Derived from Swenerton et al.[1583]

[w]Derived from Patel and Ryman.[1213a]

[x]Derived from Roth and Kirchgessner.[1375]

TABLE 8-5

Effects of Zinc Deficiency on Tissue Levels of Enzymes

Enzyme	Blood	Heart	Liver	Pancreas	Kidney	Testes
Alanine and aspartate aminotransferases[a]	No change[b](pig)					
δ-aminolevulinate dehydratase	Decrease[c]		No change[c]			
Arginase			Increase*[d]			
			Decrease*[b](pig)			
Glutamate, oxalate, and pyruvate transaminases			Increase*[e]			
Isocitrate dehydrogenase	No change[b](pig)	No change[b](pig)	Decrease*[b](pig)	No change[f]	No change[b](pig)	Increase[f]
			No change[f]		No change[g](pig)	
			No change[e]		No change[f]	
Leucine aminopeptidase		No change[b](pig)	No change[b](pig)		Decrease[b](pig)	
Ornithine transcarbamylase			Decrease[b](pig)			
Serine and threonine dehydratases			No change[d]			
Sorbitol dehydrogenase			Decrease*[e]			
Succinic dehydrogenase			No change[f]		No change[g](pig)	No change[f]
					No change[f]	
Tryptophan pyrrolase			Increase*[d]			

Conditions:

Decrease - Deficient lower than either ad libitum or restricted-fed controls.

Decrease* - Restricted-fed as well as deficient lower than ad libitum controls; difference not necessarily significant.

No change - Deficient not different from ad libitum controls.

Increase - Deficient greater than ad libitum controls.

Increase* - Restricted-fed as well as deficient greater than ad libitum controls; difference not necessarily significant.

[a]Studies done in the rat unless otherwise noted.

[b]Derived from Burch et al.[201b]

[c]Derived from Finelli et al.[473]

[d]Derived from Hsu and Anthony.[730]

[e]Derived from Roth and Kirchgessner.[1372]

[f]Derived from Prasad and Oberleas.[1279]

[g]Derived from Prasad et al.[1285]

CONCLUSIONS

1. A fairly constant supply of zinc is required by all species.

Signs of zinc deficiency develop rather quickly when zinc intake is low because very little zinc is stored in the body in a readily available form. Although a relatively large amount of zinc is stored in skin, muscle, and bone, this zinc is not available to the animal unless those tissues are being catabolized.

2. The extent to which marginal zinc deficiency occurs in humans and animals is unknown.

Without a specific test for zinc deficiency, it is difficult to determine a marginal occurrence of it. The high protein diet typical of middle and upper income Americans will probably supply close to the 15 mg zinc that is the RDA for adults. Therefore, deficiencies would not be expected in these individuals. However, people eating low protein diets are apt to receive significantly less zinc. To what extent marginal zinc deficiency may occur under these conditions is at present unknown. It is important to realize that the RDA and nutrient requirements of animals are simply the best estimates that can be made with the limited amount of data available. They will undoubtedly be modified as more is known about the factors affecting zinc requirements and zinc availability.

3. The aspects of cellular metabolism that are primarily affected by zinc deficiency are not known.

Although zinc is contained in a number of enzymes, the effect of zinc deficiency on enzymatic activity is varied, and decreased activity of known zinc-dependent enzymes does not seem to be a major factor in disturbances observed in zinc deficiency. Many of the effects observed may be a function of low food intake or lack of growth.

4. The amount of zinc occurring naturally in food is unlikely to be a health hazard.

Ingested zinc is relatively nontoxic if the diet contains adequate copper and iron; most animals appear to tolerate levels of the order of a milligram per gram. Because of the possible interference with copper and iron metabolism, animals with high zinc intakes should be monitored for signs of anemia.

CHAPTER 9

ZINC IN METALLOPROTEINS

SPECIFIC ZINC ENZYMES

The study of the specific biologic role of zinc has centered on its function in a number of zinc metalloenzymes. Several of their structures are now known on a molecular level; moreover, the three-dimensional structure as determined by X-ray diffraction has allowed some precise descriptions of the role of zinc in the mechanisms of enzymatic action. The characteristics of the zinc metalloenzymes which have been well documented and the reactions catalyzed are listed in Table 9-1. Table 9-2 lists zinc-containing enzymes for which extensive studies on the function of zinc have not been carried out. Some enzymes, such as DNA and RNA polymerase, have been reported to contain zinc when isolated, but the metal's particular functional role in these enzymes has not been delineated.

The carboxypeptidases (part of the exocrine secretion of the pancreas) are important in the C-terminal hydrolyses of peptides and proteins in the mammalian gut. Additional proteases from bacteria have been discovered to be zinc metalloenzymes. The most thoroughly studied example is thermolysin, found in an extremely heat-stable enzyme containing both calcium and zinc.[990] The Ca(II) appears to participate in the heat stability rather than in the activity, whereas Zn(II) is essential to activity and appears to function much as the Zn(II) ion in carboxypeptidase A. The specificity of thermolysin is analogous to that of the latter enzyme, except that it is an endopeptidase and does not require the terminal free-carboxyl on the substrate. The neutral protease from Bacillus subtilis has been less extensively studied, but it is known that Zn(II) is essential for activity.[1635]

TABLE 9-1

Well-Characterized Zinc Metalloenzymes[a]

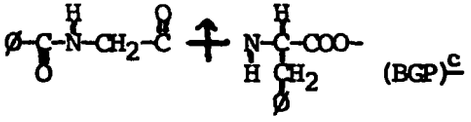
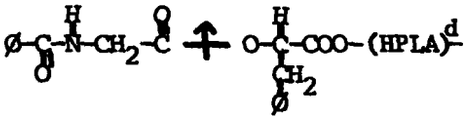
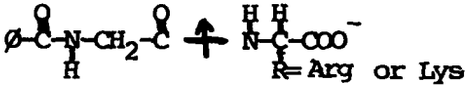
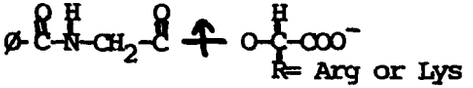
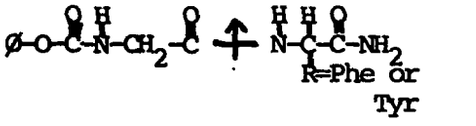
Enzyme	Molecular Weight	Zinc per Molecule	Reaction Catalyzed[b]	Reference
Carboxypeptidase A	34,500	1	 (BGP)[c]	Hartsuck and Lipscomb[644]
			 (HPLA)[d]	
Carboxypeptidase B	34,500	1	 R= Arg or Lys	Folk[484]
			 R= Arg or Lys	
Thermolysin	34,600	1; 4 Ca		Matthews et al.[990]
Neutral protease	44,700	1-2	 R=Phe or Tyr	Tsuru et al.[1635]
Leucine aminopeptidase	54,000	2	H_2N-leucine $\rightleftharpoons$ NH_2(or peptide)	Himmelhoch[706]
Carbonic anhydrase	30,000	1	$CO_2 + H_2O(OH^-) \rightleftharpoons H_2CO_3(HCO_3^-)$	Lindskog et al.[942]
			$RC(=O)OR' + H_2O \rightleftharpoons RCOOH + R'OH$	
			$RCHO + H_2O \rightleftharpoons RC(OH)_2H$	

TABLE 9-1 (Continued)

Enzyme	Molecular Weight	Zinc per Molecule	Reaction Catalyzed[b]	Reference
Alkaline phosphatase	80,000	2	$ROPO_3 + H_2O \rightleftharpoons ROH + HPO_4^{=}$ $RSPO_3 + H_2O \rightleftharpoons RSH + HPO_4^{=}$ $ROPO_2S^{=} + H_2O \rightleftharpoons ROH + HPO_3S^{=}$ $RNPO_3^{=} + H_2O \rightleftharpoons RNH_2 + HPO_4^{=}$	Reid and Wilson[1322]
Aldolase (yeast)	80,000	2	Fructose-1,6-diP ⇌ dihydroxy-acetone-P + glyceraldehyde-3P	Horecker et al.[724]
Alcohol dehydrogenase	80,000	4; 2 NAD	$CH_3CH_2OH + NAD \rightleftharpoons CH_3CHO + NADH + H^+$	Keleti[833]
Superoxide dismutase	32,000	2; 2 Cu	$O_2^- + O_2^- + 2H^+ \rightleftharpoons H_2O_2 + O_2$	Fridovich[508]
Aspartate transcarbamylase	300,000	6	$H_2N-\overset{O}{\overset{\|}{C}}-O\sim P + Asp \rightleftharpoons$ Carbamyl-Asp $+ HPO_4^{=}$	Wiley et al.[1772]
6 catalytic	subunits 33,000			
6 regulatory	subunits 17,000			

[a] See also Chlebowski and Coleman.[275a]

[b] ↑ = bond hydrolyzed

[c] Benzoylglycylphenylalanine

[d] Hippurylphenyllactic acid

TABLE 9-2

Enzymes Reported to Contain Zinc[a]

Enzyme	Source	Molecular Weight	Zinc Atoms per Molecule	Cofactor	Reference
RNA polymerase	Escherichia coli	370,000	2		Scrutton et al.[1462]
RNA polymerase	T_7 phage	110,000	2-4		Coleman (unpublished)
DNA polymerase	Escherichia coli	109,000	2		Slater et al.[1494]
Nucleotide pyrophosphatase	Rat liver		0		Corder and Lowry[316]
5'-Nucleotidase	Escherichia coli	52,000	1		Dvorak and Heppel[410]
Cyclic phosphodiesterase	Escherichia coli		0		Dvorak and Heppel[410]
Phosphomannose isomerase	Yeast	450,000	1		Gracy and Noltmann[553]
Phosphoglucomutase	Yeast		1		Hirose et al.[708]
α-D-Mannosidase	Jack bean (Canavalia ensiformis)		0		Snaith and Levvy[1518]
β-Lactamase	Bacillus cereus		0		Sabath and Finland[1388]
Protease	Snake venom	26,000	1; 2 Ca		Wagner and Prescott[1713]
5'-Adenosine monophosphate aminohydrolase	Rat muscle	290,000	2		Zielke and Suelter[1819]
Collagenase	Clostridium histolyticum	105,000	0		Seifter et al.[1470]
Neutral protease	Bacillus cereus		0		Feder and Garrett[458]
Dipeptidase	Porcine kidney	47,000	1		Campbell et al.[225]
Phospholipase C	Bacillus cereus				Ottolenghi[1184]

TABLE 9-2 (Continued)

Enzyme	Source	Molecular Weight	Zinc Atoms per Molecule	Cofactor	Reference
Dipeptidase	Mouse ascites tumor	87,000	1		Hayman and Patterson 654
α-Amylase	Bacillus subtilis	50,000	0.3 Ca		Vallee and Wacker 1665
D-glyceraldehyde-3P-dehydrogenase	Porcine muscle		3	3 NAD	Keleti 832
Lactic dehydrogenase	Rabbit muscle		0	NAD	Vallee and Wacker 1665
Malic dehydrogenase	Bovine heart	40,000	1		Harrison 639
Glutamic dehydrogenase	Bovine liver	1,000,000	2-4	0 NAD	Adelstein and Vallee 9
Transcarboxylase	Proteus shermanii	670,000	4; 2 Co	6 Biotin	Northrup and Wood 1148
Pyruvate carboxylase	Yeast	600,000	4	4 Biotin	Scrutton et al. 463
Mercaptopyruvate sulfur transferase	Escherichia coli	23,800	1		Vachek and Wood 1649
Rhodanase (sulfur transferase)	Bovine liver	37,000	2		Volini et al. 1702
5-Aminolevulinic acid dehydratase	Bovine liver	260,000	0		Wilson et al. 1787

[a] However, definitive data relating zinc to the structure or function of these enzymes are not available.

Carbonic anhydrase plays an important role in catalyzing the rate of attainment of equilibrium between carbon dioxide and the bicarbonate anion, a process involved in many important cell functions in the mammal: carbon dioxide diffusion at the alveoli, hydrochloric acid secretion in the stomach (bicarbonate anion exchange for chlorine anion) and acid secretion in the renal tubule. Carbonic anhydrase may also influence the $CO_2 \rightleftharpoons HCO_3^-$ equilibrium in the leaves of green plants. Aside from carbon dioxide transport itself, it is possible that catalysis of the $CO_2 \rightleftharpoons HCO_3^-$ equilibrium by carbonic anhydrase is involved in the general scheme of anion transport and the maintenance of electroneutrality.

Alkaline phosphatase is widely distributed in nature. It is present in bacterial cells like Escherichia coli (where it is located in the periplastic space between cell wall and cell membrane), as well as in mammalian cells such as the cells of the intestinal endothelium, and cells along the line of newly calcifying bone. Both the bacterial and mammalian enzymes are zinc metalloenzymes. The known function of alkaline phosphatase is the nonspecific hydrolysis of phosphate monoesters, but many of the enzyme's functions in the complex environment of the cell are not fully understood and it also may participate in phosphate transport and phosphate transfer reactions. Bone phosphatase is essential for bone calcification, as human mutants lacking the enzyme activity do not calcify their bones. The enzyme does participate in transferase activity in the test tube with suitable acceptors such as Tris buffer and ethanolamine. Acceptors must carry an amino function on a carbon adjacent to a carbon carrying an alcoholic hydroxyl.

The physiologic implications of alcohol dehydrogenase are obvious because of its central position in the metabolic pathway for the exogenous (and possibly endogenous) metabolite, ethanol.

Superoxide dismutase, a zinc-copper protein also known as erythrocuprein, has become the subject of recent interest because of the theory that its presence is necessary for organisms carrying out aerobic metabolism by protecting the organism from damage by the superoxide radical. This radical is the product of a number of enzyme-catalyzed reactions involving oxygen: for example, the oxidation of xanthine to urate by xanthine oxidase. Radicals as products are likely to occur since reactions involving oxygen usually proceed in one-electron steps involving free-radical intermediates because of the spin restriction applying to the addition of two electrons to molecular oxygen. The enzyme catalyzes the dismutation of the superoxide radical rapidly, limited only by the difusion of substrate.

Aldolase is a central enzyme in the pathway of anaerobic glycolysis and in organisms such as yeast, Zn(II) is an essential component although the mammalian muscle enzyme is not a metalloenzyme. In this enzyme, a specific lysyl $-NH_3^+$ group is the substitute for the zinc function.

Aspartate transcarbamylase is important in pyrimidine biosynthesis and the bacterial enzyme is the only well-characterized zinc metalloenzyme in which zinc directly and exclusively affects enzyme structure, since the metal is contained in the regulatory subunit of this enzyme and the catalytic subunit is active in the absence of metal. Zinc is involved indirectly in the regulation of enzymatic activity, since its presence is required for the proper association of regulatory and catalytic subunits to form the functioning hexamers of this allosteric enzyme.

ROLE OF ZINC AT THE MOLECULAR LEVEL

The protonatable amino acid side chains of proteins have been the obvious candidates as ligands to the zinc in metalloproteins. These chemical groups include the carboxyl groups of aspartyl and glutamyl residues, the N-terminal α-amino groups of the peptide chain, the ϵ-amino groups of lysyl residues, the imidazole nitrogens of histidyl residues -- the most likely candidates considering their chemical properties of pK_a and formation constants -- the phenolic hydroxyl groups of tyrosyl residues, the sulfhydryl groups of cysteinyl residues and the guanidino groups of arginyl residues. The -NH-group of the peptide bond has been shown in certain model systems, especially Zn(II)-peptide complexes, to form a coordinate bond with loss of its proton. Thus this group is also a potential donor in the formation of metal-protein complexes.

Of this group, only three have been identified as Zn(II) ligands in Zn(II) metalloproteins: the imidasole nitrogen, the γ-carboxyl group of glutamic acid and the sulfhydryl group of cysteine. Carboxypeptidase A coordinates the Zn(II) through the imidazole nitrogens of two histidyl residues and the γ-carboxyl group of a glutamyl residue.[644] Thermolysin, a bacterial endopeptidase similar to carboxypeptidase A, has the same three ligands to the Zn(II) ion.[990] In both proteins, the fourth coordination position of a distorted tetrahedron is occupied by a water molecule. The Zn(II) of carbonic anhydrase is coordinated to the imidazole nitrogens of three histidyl residues. A fourth coordination site is also open to solvent and occupied by a water molecule or an hydroxyl ion.[275a,303,942] Indirect evidence from nitrogen nuclear hyperfine splitting of the electron spin resonance signal of the Cu(II) derivative of alkaline

phosphatase suggests the presence of three nitrogen nuclei as ligands to the metal ion (probably imidazole nitrogens).[275a] Spectral data and amino acid analysis have established that Zn(II) binds to sulfhydryl groups in the protein metallothionein from mammalian kidney.[275a,303] Zn(II) also binds to sulfhydryl groups in alcohol dehydrogenase.[275a]

To draw conclusions about the nature of coordination sites to be found at the active centers of metalloenzymes, the large body of information that exists for small coordination complexes must be studied. In these systems, however, the ligands are flexible and free to assume bond lengths and bond angles dictated by the electron distribution in the d-orbitals of the metal ion. Bond lengths and bond angles in enzyme coordination sites, however, may be dictated as much by the stereochemistry of relatively inflexible protein ligands as the electron distribution in the metal ion's orbitals. Such inflexibility may be one of the advantages of a large protein molecule as a catalyst in that a particularly unstable configuration of amino acid residues (considered as an isolated structure) may be maintained at one site in the molecule (e.g., the active site) by the stabilizing effect of structure in the rest of the molecule. Such "strain" could radically affect the stability of particular complexes and affect the reactivity of the metal ion toward a substrate in a mixed complex formed during catalysis.

Highly purified zinc metalloenzymes for which the relative affinities of several first transition and IIB metal ions for the metal-binding site have been determined show an extraordinary preference for Zn(II) compared to Co(II), Ni(II), and Cu(II), although the sites in most cases are made up of nitrogen and oxygen ligands, which have greater affinity for the other three ions in simple systems. Model systems would suggest that all Zn(II) binding sites should contain sulfur ligands, since sulfur is known to have a particularly high affinity for Zn(II), a property that does not occur uniformly in nature.

The Zn(II) ion does not appear to be necessary for the synthesis of the apoprotein nor does it have to be present to assure the presence of the high affinity Zn(II) binding site. The Zn(II) enzyme, alkaline phosphatase, has been thoroughly examined in this regard. Escherichia coli grown in a zinc-free medium derepressed for alkaline phosphatase will produce a zinc-free inactive apoalkaline phosphatase with physicochemical characteristics identical to the apoenzyme produced by removal of the metal with chelating agents.[275a] The apoenzyme synthesized by the Zn(II)-deficient organisms can be completely reactivated by the addition of Zn(II). The organism can synthesize enzymes containing Co(II), Cu(II), and Cd(II) if these metal ions are added to the zinc-free medium, but Zn(II) will overpower the other three metals at relative concentrations much below those expected for competititve binding to model coordination complexes. This dominance could be related to the fact that most of the protein binding sites have highly distorted ligand geometry, a function of the three-dimensional structure of proteins. The first transition metals with unfilled d-shells generally derive significant stabilization of their complexes from the ligand field stabilization energy, provided the geometric arrangement of the ligands is such that low energy orbitals exist that can be occupied and higher energy orbitals are left vacant or half-occupied. If the ligands are prevented from assuming the ideal geometry that maximizes the splitting of the orbital energy levels (as may occur because of restraints in the protein structure), the metal complex will be destabilized in relation to the usual models. However, zinc with a filled d-shell and no field stabilization energy from the ligand may accommodate the distorted geometry without destabilization resulting.

It has been speculated that zinc operates in its capacity as a Lewis acid and withdraws electrons from certain groups of the substrate in mixed enzyme-metal-substrate complexes. The open coordination sites in carboxypeptidase A,

thermolysin, and carbonic anhydrase clearly suggest this possibility. Only in the case of carboxypeptidase A has the structure of an enzyme-metal-substrate complex been determined. The X-ray structure of the crystalline glycyl-L-tyrosine complex with carboxypeptidase A shows the Zn(II) ion to be coordinated to the carbonyl oxygen of the susceptible peptide bond which is placed in the position of the coordinated water molecule in the free enzyme.[644] Mechanisms proposed for carbonic anhydrase suggest coordination between Zn(II) and the oxygen of an active hydroxyl ion in hydration or the oxygen of bicarbonate in the dehydration reaction, but no direct proof is available.[275a] Whether a simple Lewis acid function applies in all instances or even in all hydrolysis or hydration reactions catalyzed by zinc metalloenzymes is not clear. Structural roles for Zn(II) have been suggested, but evidence has been difficult to obtain. In aspartate transcarbamylase the Zn(II) is not at the active site, but seems to participate primarily in the structure of the regulatory subunit.[1772]

EVOLUTION OF ZINC ENZYMES

If one of the major functions of zinc in the biosphere is its fundamental catalytic role in metalloenzymes, then an important question is, how universal among the various phyla is it that the catalysts for these particular reactions are zinc metalloproteins? Is the evolution of a given zinc metalloenzyme a relatively restricted event, or have these zinc macromolecular catalysts existed over long periods of evolution? A great amount of phylogenetic information has been gathered on some of these zinc enzymes from isolating and characterizing the enzymes (listed in Table 9-1) from plants, and primitive and higher animals all the way up to primates, including man. The sources from which the enzymes were obtained, the molecular weight, and the zinc content are given in Table 9-3. Usually if an enzyme is a zinc enzyme

in one species, it will contain zinc in another; for many enzymes the zinc is maintained in the enzyme from bacteria to man. Thus in an enzyme like carbonic anhydrase, the active site of zinc is present in the enzymes of the elasmobranch, a species that split off from the mainstream of animal evolution approximately 350 million years before the evolution of mammals. Therefore some zinc enzymes are under little evolutionary pressure to change.

However, there are notable exceptions. The aldolase of yeast is a zinc enzyme, whereas the aldolase of the mammalian muscle is definitely not. An ε-amino group of a lysyl residue of the protein provides a complete reaction through the formation of a ketimine intermediate without the assistance of a metal ion. Another notable substitution occurs within the enzyme superoxide dismutase: the bacterial enzyme is a manganese metalloprotein, but the mammalian enzyme is a zinc-copper protein.

OTHER POSSIBLE BIOLOGIC FUNCTIONS OF ZINC

Whereas the enzymatic functions of zinc outlined above form a specific and fundamental basis for the biologic function of zinc, a number of complex syndromes have been described in animals as a result of zinc deficiency.[1652] In many of these syndromes, however, it is very difficult to ascribe the pathologic findings (both chemical and morphologic) to the malfunction of all or some of the zinc enzymes described above. This may relate to the difficulty of predicting the chemical and morphologic changes responsible for disrupting a single or limited number of enzymatic steps in a complex and interrelated series of metabolic pathways. For example, a disorder similar to cirrhosis of the liver appears in zinc-deficient pigs and a relationship to alcohol dehydrogenase might be postulated. It has been found that serum zinc content of alcoholic humans with cirrhosis has been low and urinary zinc excretion has been high.

However, some features of zinc deficiency syndromes reflect unknown biologic functions of zinc. One example is the well-characterized zinc deficiency that occurs in Euglena gracilis.[1710] In zinc-deficient media, these organisms fail to grow well, and undergo peculiar morphologic changes. Chemical analyses have shown that the concentration of DNA per cell doubles, whereas protein and RNA synthesis are radically depressed. In addition, a derangement of the relative concentration of other metals and a striking increase in acid-insoluble polyphosphate occur.[1710] It has been suggested that zinc participates either in the structure and function of nucleic acids or in protein synthesis, through effects on ribosomal structure or on specific enzymes involved in protein synthesis. Indeed, it has been shown that RNA isolated from diverse biologic sources does contain zinc[1712] and that added zinc and other metal ions significantly affect RNA structure, as revealed in melting curves.[518]

Zinc may have important enzymatic functions of which we are not yet fully aware. For example, isolated samples of DNA and RNA polymerase contain zinc (Table 9-2). If zinc were essential to proper transcription in Euglena, the chemical findings reported would be consistent with the activity necessary to that function. However, finding zinc in a given biologic specimen is not in itself sufficient evidence for function, because zinc is a ubiquitous element in biologic fluids, second only to iron in relative concentration among the transition elements. More direct relationships between the contained metal and enzymatic activity must be demonstrated. Zinc has been clearly proved to participate in the structures and mechanisms of action of many enzymes. But it is possible that zinc also functions in maintaining the ionic environment necessary for certain biologic processes or in the maintenance of the structure of certain nonenzymatic proteins. For example, the cadmium-containing protein metallothionein has been isolated from kidney,[807] and shown to contain substantial amounts of zinc. The biologic function of this protein is not known at present.

TABLE 9-3

Comparative Biochemistry of Zinc Metalloenzymes

Enzyme	Source	Molecular Weight	Zinc Content, gram atom/mole[a]	Reference
Carboxypeptidase A	Bovine pancreas	34,500	1	Vallee and Neurath[1661]
	Porcine pancreas	34,000	1	Folk and Schirmer[486]
	Spiny dogfish pancreas	35,000	contains zinc	Lacko and Neurath[889]
Carboxypeptidase B	Bovine pancreas	34,000	1	Wintersberger et al.[1792]
	Porcine pancreas	34,300	1	Folk and Gladner[485]
	Spiny dogfish pancreas	35,000-37,000	contains zinc	Prahl and Neurath[1270]
Carbonic anhydrase[a]	Human red cell	30,000[a]	1	Armstrong et al.[33]
	Rhesus monkey red cell	30,000	1	Duff and Coleman[405]
	Bovine red cell	30,000	1	Keilin and Mann[831]
	Equine red cell	30,000	1	Furth[517]
	Porcine red cell	30,000	1	Tanis et al.[1596]
	Canine red cell	30,000	1	Byvoet and Gotti[214]
	Guinea pig red cell	30,000	1	Carter and Parsons[243]
	Elasmobranch red cell			Maynard and Coleman[997]
	Bull shark	40,000	1	
	Tiger shark	40,000	1	
	Cuttlefish gill *Sepia officinalis*		contains zinc, but inhibited by chelating agents	Addink[6]

TABLE 9-3 (Continued)

Enzyme	Source	Molecular Weight	Zinc Content, gram atom/mole[a]	Reference
Carbonic anhydrase[a]	Parsley leaves	180,000 (29,000 subunit)	1 29,000 g	Tobin[1624]
	Neisseria sicca	28,000	1	Brundell et al.[180]
Alcohol dehydrogenase	Human liver	87,000	2-4	von Wartburg et al.[1706]
	Horse liver	84,000	2-4	Vallee and Hoch[1659]
	Yeast	150,000	4	Vallee and Hoch[1658]
Alkaline phosphatase	Human placenta	125,000	0.15%	Vallee and Hoch[1658]
	Human leukocytes		0.15%	Trubowitz et al.[1629]
	Human bone		zinc enzyme by histochemical staining with zinc chelate	Bourne[142]
	Calf intestine	100,000	0.20%	Mathies[985]
	Escherichia coli	80,000-89,000	2-4	Plocke et al.[1254]; Applebury and Coleman[48]
	Bacillus licheniformis	121,000	no data	Hulett-Cowling and Campbell[747]
	Bacillus subtilis	100,000	2	Yoshizumi and Coleman[1812]
Aldolase (type I)	Rabbit muscle	160,000	no zinc	Horecker et al.[724]
	Other mammalian muscle	160,000	no zinc	
Aldolase (type II)	Yeast	80,000	2	Richards and Rutter[1345]

TABLE 9-3 (Continued)

Enzyme	Source	Molecular Weight	Zinc Content, gram atom/mole[a]	Reference
Aldolase (type II)	Ascomycetes niger	—	contains zinc	Jagannathan et al.[789]
	C. utilis	—	contains zinc	Kowal et al.[872]
Superoxide dismutase		Both of two sub-units		
	Human red cell	32,000	2 plus 2 copper	Mann and Keilin[974]
	Bovine red cell	32,000	2 plus 2 copper	Fridovich[508]
	Bovine liver	32,500	2 plus 2 copper	Weser et al.[1756]
	Yeast	31,200	2 plus 2 copper	Weser et al.[1756]
	Neurospora crassa	31,000	2 plus 2 copper	Misra and Fridovich[1070]
	Escherichia coli	40,000	2 manganese no zinc or copper	Keele et al.[828]
	Streptococcus mutans	40,250	2 manganese no zinc or copper	Vance et al.[1675]

[a] Unless otherwise noted.

[b] Mammalian erythrocyte carbonic anhydrases all have molecular weights near 30,000. Actual experimental data vary from 28,000-31,000.

CHAPTER 10

CLINICAL ASPECTS OF ZINC METABOLISM

ABNORMALITIES OF ZINC METABOLISM IN DISEASE

Liver Disease

Zinc loss is a common accompaniment of all acute and chronic liver disease. Decreased concentration of zinc in serum and liver tissue and the increased excretion of zinc in the urine have been reported in patients with alcoholic cirrhosis of the liver.[610,1568,1666,1667,1668] Prasad et al.[1273] found decreased zinc in red blood cells of cirrhotic subjects. Oral administration of zinc sulfate was reported[1568] to improve some liver functions, but these studies were uncontrolled and attempts to confirm their results have not been successful.[1666,1667] In chronic alcoholism, patients commonly exhibit serum zinc concentrations two standard deviations below the normal mean, elevated urinary zinc excretion, and increased clearance of renal zinc.[1568] Urinary zinc excretion returned to normal in some patients without cirrhosis who abstained from alcohol for 1-2 wk.[1568] But in patients with postalcoholic cirrhosis the changes persist. Administration of alcohol to normal subjects has been associated with a slight increase in urinary zinc excretion, but it is difficult to predict whether such an increase may occur.[1564]

The zincuria of alcoholism may be somewhat similar to the renal defect for magnesium found in alcoholics.[1568] Prolonged and excessive intake

of alcohol with insufficient dietary intake of protein and other nutrients may produce primary renal dysfunction with excessive excretion of zinc and/or magnesium. Use of intravenous zinc-65 suggested that the hyperzincuria in cirrhotic patients was not caused by a specific renal defect;[1567] a more general abnormality of cellular zinc metabolism was postulated.

Rats with cirrhosis exhibited reduced serum and liver zinc levels.[810] Laboratory models of hepatic cirrhosis induced by carbon tetrachloride and other agents have also been used to study the role of zinc in liver abnormalities. Liver damaged by carbon tetrachloride has been used to study other manifestations of liver pathology related to zinc. Voigt and Saldeen[1701] showed that water-soluble zinc salts given parenterally inhibit liver damage produced by manganese in golden hamsters and carbon tetrachloride in rats. A histochemical study[1395] demonstrated that mice with hepatic damage from carbon tetrachloride had much less pronounced enzyme changes when treated with zinc beforehand to protect them. In animals treated simultaneously with carbon tetrachloride and zinc chloride, alkaline phosphatase increased in newly formed tissue in the peripheral zone of the damaged liver. However, no difference in activity between zinc metalloenzymes or zinc-dependent enzymes was found.[1396] The uptake of zinc-65 in rat liver damaged by carbon tetrachloride increased; uptake was greater in animals with damaged livers than in controls.[1809] Zinc was taken up by the parenchymal cells but not by the Kupffer cells.[1396] However, even after acute carbon tetrachloride damage, uptake of zinc-65 was still rapid,[89a] suggesting that any alteration in zinc-65 uptake in damaged rat liver requires time following the insult to be apparent.

In all patients with infectious hepatitis, serum concentrations of total zinc decreased early in the acute phase of the viral disease and then rose to normal as the illness subsided;[683] diffusible serum zinc concentrations were elevated early in the acute phase of the disease and fell to normal as the disease subsided.[683] Serum changes were always accompanied by a significant zincuria during the early acute phase of the illness and by a return to normal as the illness subsided.

Little about the zincuria or the levels of plasma zinc of hepatitis patients has been agreed upon; some investigators discovered no change from normal urinary zinc excretion but a lowering of plasma zinc (which eventually returned to normal),[819] while others noted variability in urinary excretion and plasma concentrations of zinc.[361,608,809] The disagreement may be related either to the failure to specify the stage of the illness at which the patients were studied or to differences in methodology. Studies carried out late in the disease may have missed the elevated urinary zinc excretion and lowered serum zinc levels observed in the acute phase.

Decreased concentrations of total plasma zinc and increased urinary zinc excretion have been observed in patients with several forms of liver disease.[610, 683,819] Attempts have been made to correlate abnormal levels of plasma zinc with the severity of the hepatic disease with moderate success.[661,809] In patients with acute viral hepatitis, the lowest serum total zinc concentrations and the highest levels of diffusible serum zinc and urinary zinc excretion were observed in patients hospitalized because of the severity of their disease.[683]

Normally, there are two classes of zinc ligands in serum. Macromolecular zinc ligands, including albumin and α_2-macroglobulin, exemplify one class;[537,538,668] micromolecular zinc ligands, including amino acids and species such as porphyrins and peptides, exemplify the other.[537,668] The equilibrium of these ligands in serum depends upon their respective concentrations of albumin and amino acids, particularly levels of histidine and cysteine.[537,538,668] In vitro, the amount of ultrafilterable or diffusible zinc has been shown to increase as the concentration of these amino acids increases. Exchangeable zinc is bound mainly to albumin, and the affinity of zinc for albumin is altered sharply in favor of the amino acids histidine and cysteine if their concentration in serum is elevated or if albumin concentration is decreased.[537,538,668] However, urinary zinc excretion is related primarily to the amount of micromolecular-liganded zinc complexes because they readily pass the renal glomerulus and are excreted in urine. Indeed, oral administration of histidine to humans has produced significant hyperzincuria and hypozincemia.[673,679]

In general, significant decreases in plasma albumin or globulin do not occur in acute viral hepatitis.[1481a] No such decreases were observed in the patients studied by Henkin and Smith.[683] However, the concentration of some amino acids in plasma have

been reported to increase by 20%[460a] and increased urinary excretion of amino acids such as histidine, lysine, and glutamine has been observed.[1723a] These amino acids also may be released into the circulation during the destruction of liver cells that accompanies severe liver disease.[683] Thus, the increased concentration of plasma and urinary free amino acids observed in hepatitis might be associated with shifts in the binding of zinc from macromolecular to micromolecular ligands and consequent increases in diffusible zinc. These changes would account for the significant zincuria and liver losses of zinc observed in this disease.

Another mechanism by which diffusible zinc might be increased during hepatitis is by reducing the affinity of the macromolecular zinc ligands for zinc. In those patients observed by Henkin and Smith, serum diffusible zinc varied directly with bilirubin concentration.[683] When calculated separately, the correlation between plasma bilirubin and diffusible zinc in patients with bilirubin concentrations greater than 6 mg/100 ml (correlation coefficient = 0.949) was greater than that in patients with bilirubin concentrations less than 6 mg/100 ml (correlation coeficient = 0.605). Whereas the correlations in the entire group and in each of the two subgroups were all highly significant (probability < 0.001), the higher correlation coefficient suggests a closer relationship between bilirubin and diffusible zinc at higher bilirubin concentrations. Nevertheless, the normal concentration of albumin in plasma (650 μM) is so much greater than that of zinc (15 μM) or bilirubin (1 μM), that even

in the event of severe pathologic increases in bilirubin,[478a,762b,1652,1809a] the displacement of zinc by bilirubin from a common binding site cannot account for the observed increase in the concentration of diffusible zinc.

One attempt to clarify this complex relationship was made by Lindeman and Baxter, who suggested that alterations in serum binding of zinc-65 were present in patients with cirrhosis.[933a] Schecter et al.[1420a] studied the distribution of serum zinc between albumin and α_2-macroglobulin in patients with decompensated hepatic cirrhosis. Not only did those patients with cirrhosis exhibit lower than normal zinc levels, but a greater proportion of the zinc in their serum (about 50%) was associated with α_2-macroglobulin, an amount significantly higher than percentages found in normal subjects (between 20-30%). Schecter et al.[1420a] also noted that albumin-bound zinc in serum was lower than that of normals, as expected.[809,1715a] Decreased levels of zinc have also been found in the serum, red and white blood cells,[504b] and hair[1285a] of cirrhotic patients. Increased urinary zinc excretion and a decreased total body zinc pool, was observed. All of these symptoms indicate that patients with decompensated hepatic cirrhosis exhibit a total body loss of zinc.

Other Gastrointestinal Disorders and Malabsorption

Various abnormalities of zinc in blood have been reported in patients with gastrointestinal disorders. Lower than normal levels of serum zinc have been reported in patients with kwashiorkor,[630a,1412,1504] although hair zinc levels are elevated above normal.[867a] Lower than normal levels of zinc in blood have been reported in patients with regional enteritis;[1404a ,1522] several malabsorptive

states;[998a,1715a] acute dysentery;[140] gastric ulcers complicated by pyloric stenosis;[1563] and achylia gastrica.[1802] However, no abnormal hematic zinc levels were found in patients with acute gastrointestinal hemorrhage[361] or following partial distal gastrectomy.[972a] Indeed, experimental production of gastric ulcers in dogs revealed decreased fecal zinc excretion and presumed zinc retention in tissues.[1203a] Increases in whole blood zinc in patients with chronic pancreatitis have been reported, the result of decreased plasma zinc and increased red blood cell zinc.[1817a] Zinc deficiency may affect exocrine activity of the pancreas. The metal has been shown to be an important component of pancreatic carboxypeptidase,[1661] and the activity of this enzyme was reduced in zinc-deficient rats.[1066]

Hypogonadal Dwarfism Syndrome

Dietary zinc deficiency has been associated with clinical symptoms in humans. Prasad et al. reported on a group of Iranian men whose diet consisted almost exclusively of bread and beans.[1276] Their deficiency was related not only to the low level of zinc and protein in their diet, but the presence of phytate in the bread. Phytate tends to decrease the intestinal absorption of endogenous dietary zinc.[1276,1277,1278,1287,1412] Symptoms of deficiency in these patients were decreased zinc in plasma, red blood cells and hair; decreased urinary zinc excretion; rapid turnover rate of zinc-65 with decreased 24-h exchangeable pool; and decreased excretion of zinc-65 in feces and urine.

Iron deficiency anemia, dwarfism, hypogonadism, hepatosplenomegaly, depression of adrenocorticotropin (ACTH) production, and increased sensitivity to insulin[1409] was also found in a group of Egyptian adult males on a low-protein diet[1412] similar to that observed in the previously reported Iranian men. Although the Egyptian boys studied were infested with liver schistosomes, they excreted less urinary zinc than did normal subjects; this tendency contrasts with the zincuria seen in

patients with cirrhosis of the liver on zinc-adequate diets. This observation suggested that these subjects, taking a diet which was apparently low in zinc, conserved zinc by reducing its excretion in urine.

Anemia, hepatosplenomegaly, growth retardation and lower than normal levels of zinc in plasma were also observed in preadolescent Iranian children.[436a] The basis for this syndrome was also considered to be the large amounts of phytate and fiber present in their diet. Dwarfism, hypogonadism and alterations in zinc metabolism have also been reported to occur in the U.S. without association with the dietary intake of large amounts of phytate or fiber.[214a]

Zinc salts and a good animal protein diet were fed to some Egyptian boys. All symptoms of the syndrome were reported to diminish or disappear, and the boys gained weight. The level of zinc in body fluids and hair were reported to return to normal levels. One patient only on an iron supplementation did not show these changes. After their plasma zinc had become normal, five patients returned to their native villages and resumed their original diets; their plasma zinc levels gradually decreased, reinforcing the conclusion that the described syndrome was caused by zinc deficiency. Ronaghy et al. reported that administration of zinc supplements to malnourished Iranian school boys improved their skeletal growth.[1363]

Several investigators have doubted that zinc deficiency alone produced this syndrome. The reported symptoms of these patients coexisted not only with zinc deficiency, but also with protein deficiency, and were manifested by depressed levels of serum albumin and plasma protein.[248] Thus, Coughey[248] considered the role of zinc deficiency overemphasized in these patients. In the only controlled clinical trial carried out in this patient group, a double-blind study of the administration of zinc sulfate and placebo indicated that no differences could be shown between the treatment groups.[1410] Plasma and erythrocyte zinc levels are below normal in children suffering from protein calorie malnutrition;[885] these levels are lower in kwashiokor than they are in marasmus.[885] Hair zinc in growth-retarded Iranian[248] and Egyptian[248,1558] hypogonadal patients was below normal,[248]

although hair zinc levels in patients with either kwashiorkor or marasmus have been found to be similar[25a] to or appreciably higher than in normal subjects.[867a] In these latter patients plasma or erythrocyte zinc is lower than in normals or in patients with dietary zinc deficiency. These differences may be useful in differentiating between patients with abnormalities in zinc metabolism caused by dietary deficiency from those with protein calorie malnutrition.

Some Egyptian patients with hypogonadism and growth retardation have attained sexual maturity and a normal stature without treatment or change in their low plasma zinc level. Thus the importance of the plasma zinc level as a reflection of zinc deficiency may be limited. Whether zinc is the sole factor limiting growth and development in these Iranian and Egyptian adolescents has also been questioned. A debate about the limited[249,250] versus sole[1281] importance of zinc in growth and development in these patients has continued over the past few years. Although this problem has not yet been resolved, it is clear that zinc deficiency itself can be a restrictive factor in growth and development.[612] That zinc deficiency can alter other biologic systems and offer an environment in which other pathologic processes may occur is readily apparent.[609,1282,1331] In humans several pathologic processes may accompany zinc deficiency, making the issue of zinc deficiency per se difficult to define and the effects of treatment difficult to evaluate.

Acrodermatitis Enteropathica

Acrodermatitis enteropathica is a/ rare autosomal recessive disorder characterized by pathologic changes in several organ systems. Skin changes include alopecia totalis, paronychia, and bullous-pustular dermatitis of the extremities of oral, anal, and genital areas. Ophthalmic manifestations include blepharitis, conjunctivitis, photophobia and corneal opacities. Gastrointestinal manifestations include severe, often chronic diarrhea, malabsorption, steatorrhea

and lactose intolerance. Tremor has been observed, along with occasional cerebellar ataxia, emotional lability, and irritability. Infections caused by Candida albicans are frequent, as are retarded growth and hypogonadism. The disorder, described by Danbolt and Closs,[354c] and the clinical changes observed since, have been defined and reviewed.[440b,504c,802a,1232a, 1355a,1572a,1755a]

The disorder, although rare, may be observed more frequently in infants of Italian, Armenian, or Iranian origin, and usually develops after weaning from breast feeding.

Diiodohydroxyquin therapy has been used with some success in these patients since 1953,[384a] but neither the mechanism of the drug's action nor the pathologic basis for this disease was amenable to study until recently.

In 1973, decreased levels of serum zinc were noted in an infant with acrodermatitis enteropathica, and after oral treatment with zinc sulfate a complete remission of symptoms occurred.[77a,1086] After inadvertent omission of zinc sulfate from the patient's treatment, the symptoms of the disease recurred; but they remitted again following reinstitution of zinc sulfate. The dramatic alleviation of this condition with oral zinc sulfate has been confirmed.[1037b,1129a,1266a]

Reasons for the changes observed in these infants after therapy are not clear. That changes developed after weaning suggests either that infants received too little dietary zinc following weaning[620a] or that absorption of zinc was impaired. Because very small amounts of zinc added to the diet appear to correct abnormalities, it is rather unlikely that a specific dietary lack of zinc per se produces symptoms of acrodermatitis enteropathica.[672b,1129a] That treatment with diiodohydroxyquin is effective might be related to the formation of an absorbable zinc chelate.[1085b] Human milk therapy may be helpful for similar reasons.[950b]

Although there may be a significant abnormality in the manner by which orally administered zinc-65 is absorbed,[950a,950b] diverse data[515a,672b] suggest that malabsorption cannot be the primary defect in acrodermatitis enteropathica. Although the nature of the defect is not clear, the lack of an appropriate factor in gastrointestinal transport corrected by administration of even small amounts of oral zinc is an attractive hypothesis, because zinc binding and transport after absorption appear to be intact.[672b] Such a zinc-binding protein has been isolated from rat jejunal mucosa,[873] although its role in active transport across the gut mucosa has not been firmly documented.

Blood Dyscrasias

Plasma zinc was found to be below normal in pernicious anemia,[1369,1802] chronic lymphocytic leukemia,[1802] multiple myeloma,[1802] Hodgkin's disease,[58] and various other types of anemias.[1802] In Hodgkin's disease, whole blood zinc also is diminished.[58] After vitamin B_{12} therapy, the zinc serum level returned to normal in patients with pernicious anemia.[1802]

Lower than normal levels of serum zinc have also been observed in some patients with thalassemia[1275] and sickle-cell anemia,[158a,1287a] It has been suggested that zinc may counteract the deleterious effect of calcium on the red blood cell membrane. In uncontrolled studies administration of exogenous zinc has been claimed to improve the clinical status of patients with sickle cell anemia, assist in the healing of their peripheral ulcers and improve their clinical condition in general. Controlled clinical trials will be of importance to verify these provocative early findings.

Elevated plasma zinc concentration was observed in patients with eosinophilia,[1802]

lymphocytic lymphoma, severe untreated megaloblastic anemia, and multiple myeloma.[58] These latter results in patients with multiple myeloma are at variance with those noted by other investigators,[1586,1589] and make it difficult to sort out the changes that occur in blood zinc levels in this condition. In megaloblastic anemia, treatment with folic acid returned zinc levels to normal after a few days.[58] No changes in the plasma zinc level were observed in six patients with polycythemia vera or in nine patients with acute myelogenous leukemia.[1367]

Erythrocyte and plasma zinc were found to be reduced in Hodgkin's disease.[1367] Valberg et al.[1650] found an increase in the number of erythrocytes containing zinc in Hodgkin's disease, chronic myeloid and lymphatic leukemia, and in multiple myeloma, an observation contrasting with Auerbach's results.[58] The discrepancy may have been caused by different diagnostic criteria and whether patients were studied before or during therapy. The erythrocyte zinc of the leukemic subjects was higher than in the controls,[377,505] except in patients with acute granulocytic leukemia.[505] High erythrocyte zinc levels also were found in patients with untreated pernicious anemia,[1592, 1802] and pernicious anemia in relapse,[1592] although the degree of elevation did not correlate with the severity of the disease. Elevated erythocyte zinc values were also reported in patients with lymphoma and multiple myeloma,[1369] in contrast with lowered values reported by others.[1367] In megaloblastic anemia, high zinc levels in erythrocytes have been found to decrease with therapy,[505] but elevated zinc values have not always been found in megaloblastic or pernicious anemia.[1369]

In some cases, erythocyte zinc content correlated with the mean corpuscular volume of the erythrocytes, as it did in cases of polycythemia rubra vera and myeloid metaplasia.[505] However, this correlation was not observed in leukemic patients.[505] Talbot and Ross[1592] attributed the greater amounts of zinc in erythrocytes to the increased carbonic anhydrase that they found in patients with these disorders.

Zinc in leukocytes of patients with lymphatic leukemia has been reported to decrease as the number of leukocytes increased and the disease process worsened. This inverse relationship is particularly common in patients with a majority of premature forms of cells in their peripheral smear.[377] In 56 patients with chronic lymphocytic and granulocytic leukemia, or with acute lymphocytic, monocytic, and granulocytic leukemias or myeloid metaplasia, values for zinc in leukocytes were less than normal in all groups. The lowest values occurred in subjects with chronic and acute lymphocytic leukemia. No correlation between zinc in leukocytes and total or differential leukocyte number could be established.[505,1802] In patients with chronic granulocytic leukemia, zinc in leukocytes rose in response to treatment.[505] However, Frischauf et al.[510] did not find an essential difference in the zinc content in leukocytes of leukemia patients.

High concentrations of zinc in leukocytes were found in patients with predominantly lymphoblastic forms of leukemias, whereas in lymphosarcoma, Hodgkin's disease, lymphoma, and nonlymphomatous undifferentiated carcinoma, the level of zinc was close to the normal range.[237]

The zinc content in granulocytes was studied in bone marrow and peripheral blood by a histochemical technique and results were graded on a semiquantative scale.[1589] Decreased zinc content was demonstrated in acute and chronic myelocytic leukemia, plasmacytoma, and Hodgkin's disease. During the remission of leukemia the zinc content increased, but it was still below normal.[1589] Rising zinc values were observed in lymphocytic leukemia.[903,1587] In patients with myelosclerosis, the zinc content of bone marrow granulocytes was elevated.[1589] In four

cases of chronic lymphocytic leukemia,[1587] increased zinc levels were observed.

Szmigielski[1588] suggested that the amount of zinc protein in granulocytes decreased in leukemic patients. Free protoporphyrin can be demonstrated in leukemic cells, but it cannot be found in normal granulocytes; he therefore hypothesized that protein synthesis with a protoporphyrin prosthetic group was inhibited in leukemic cells. He related the decrease in zinc content to the increase of free protoporphyrin in leukemic cells, but it did not appear that the protein which regulated the maturity of granulocytes contained a zinc-protoporphyrin-prosthetic group. Decreased zinc in granulocytes of patients with acute and chronic myeloid leukemia was one of the most constant signs of the disturbances in leukemic cells.[903] As so often in zinc metabolism, the meaning of these differing observations is not immediately apparent.

Delves *et al.*[373a] reported that the ratio of plasma copper to zinc was elevated in children with untreated leukemia. This ratio was useful in evaluating patients' responses to therapy.

Other Cancers

Numerous studies of the role of or the change in serum zinc in malignancies have been performed, but the meaning of these findings is obscure. The behavior of zinc in malignancy may depend upon the site, nature and state of the cancer as well as the nature of the treatment.

Davies et al.[361] suggested that decreased zinc levels in serum of patients with carcinoma of the bronchus are one of the most constant characteristics of the disease because they remain in normal ranges in all other pulmonary illnesses. The constancy of these decreased levels has been contradicted.[361,1802] Lowered zinc plasma levels have been reported in patients with carcinoma of the colon.[361] Wolff,[1802] who studied 45 patients with carcinoma of different etiologies and sites, also found decreased serum zinc levels.[361] After studying 49 cases, Davies et al. did not find significantly lower serum zinc values in patients with carcinomas than in the control group, except for those with carcinoma of the bronchus and colon. X-ray therapy also has been said to produce an immediate increase in serum zinc values.[1802]

More recently, patients with primary osteosarcoma had elevated concentrations of serum zinc,[474a] a finding previously demonstrated.[800a] However, Fisher et al.[474a] also noted that patients with osteosarcoma with metastases had depressed serum zinc.

It has been suggested that the level of zinc in cancerous organs was higher than in normal tissue, although the tumors themselves may exhibit decreased zinc content depending on the phase of the disease.[813,1241] Serum zinc levels have been elevated in some forms of leukemia,[7] and lower than normal levels of zinc were found in leukocytes of patients with various acute and chronic leukemias.[505]

Urinary zinc was reported to be three times as high in patients with various malignancies as in normal subjects, and their urinary excretion of molybdenum was also decreased; a zinc:molybdenum ratio higher than 300:1 was considered a manifestation of cancer.[1241] An increase of the zinc:molybdenum ratio in urine was observed during the progression of the malignancy.[1650]

A small increase of zinc in 30% of patients with carcinoma was observed in erythrocytes,[1650] whereas zinc in the leukocytes of patients with different types of carcinoma showed a decrease even in very early stages of the disease.[184,185] This change has been suggested as a useful early test of the disease. A decrease of plasma zinc even as an early prognostic of cancer was suggested after analyzing the zinc content of blood granulocytes of 50 patients.[1588] The granulocyte zinc content in cancer of the skin was reported to decrease in advanced stages of the malignancy.[874]

The content of zinc in carcinoma of the prostate is of special interest because this organ normally contains the highest amount of zinc in any soft tissue in the body. In prostate, the level of zinc in malignant cells was much lower than in normal prostatic tissue.[1586] As in prostatic carcinoma, zinc content was increased rather than decreased in hyperplastic prostate glands.

Cystic Fibrosis (CF) of the Pancreas

Patients with CF exhibit higher levels of various electrolytes--particularly sodium and potassium in eccrine sweat--than do normal subjects, and this phenomenon has been useful in diagnosing the condition. However, zinc levels in hair and nails were reported to be lower than in comparable controls, although actual levels were not given.[868] Mean zinc concentration in cerumen was 16 times higher in controls than in patients with CF.[151] Pancreatic zinc concentrations in patients with CF were significantly lower than those of controls. The mean zinc concentration in the pancreases

from 17 controls was 193 ± 94 μg zinc, whereas mean concentrations for 35 patients with CF was only 77 ± 73 μg zinc/g dry pancreas. Lower levels were associated with the more severe form of the disease. Duodenal fluid from a patient with CF contained almost no zinc.[868] Differences among cerumen, hair and nails, which have lower than normal zinc levels, and sweat and saliva, which have been reported to contain higher levels, do not yield to simple interpretation.

Of further interest is the relative sterility of males and the reduced fertility of women with CF. Decreased gonadal function is a common accompaniment of zinc deficiency. Yet zinc levels in semen from men with CF are reportedly higher than in corresponding controls,[1385] because the ejaculate of CF patients contains mainly fluid from the prostate with little or no fluid from the testis or epididymus.

Infectious Processes

Zinc in serum and urine of subjects in whom experimental viral and bacterial infections have been induced has been carefully studied.[1219] Under controlled conditions, subjects were infected with viral and bacterial agents. Serum zinc levels decreased rapidly after the onset of the viremia or bacteremia, but before fever was measured.[1219] The zinc decreased throughout the course of the infectious process and was accompanied by a significant zincuria.

These changes have been related to an alteration of body zinc pools by a leukocytic endogenous mediator (LEM), a heat-labile protein released from sensitized polymorphonuclear leukocytes. LEM appears to produce a prompt hypozincemia and a concomitant increase in hepatic zinc.[94a,94b,94c, 414a,720a,814a,1217a,1217b,1219,1219a,1285a]

Lower than normal levels of zinc in blood have been observed in patients with typhus and acute dysentery.[140]

Changes in serum and urinary zinc following infection may reflect a natural body defense mechanism.[1748] Because zinc is necessary to the growth of viral or bacterial agents, decreased hematic zinc could represent a significant factor in the virostatic and bacteriostatic processes of the body.[1748] Decreased serum zinc and increased serum copper and iron are thought to be changes that occur in response to invasion by viruses and bacteria; these alterations may be part of the normally active homeostatic mechanism of the body.[1748] It is curious, however, that zinc was able to display antiviral activity at the nontoxic concentration of 0.1 μM in tissue culture of viruses.[870] The metal inhibited formation of infectious virions at any stage of the replication cycle.[870] Zinc also inhibited the cleavage of precursor molecules. Out of 11 other metals, including cadmium, copper, cobalt, mercury, molybdenum and nickel, only zinc had these antiviral effects at nontoxic levels of the metal.[871]

In vitro data suggest that at higher concentrations zinc markedly inhibits the cleavage of polio and encephalomyocarditis virus polypeptides in relation to rhinoviruses.[871]

Drug Metabolism

Various drugs alter zinc metabolism in man and animals. In the rat, *N*-methyl-*N*-nitrosourea results in retinal atrophy, cataracts, and significantly increased zinc content in the eyes.[877] However, administration of 6-azauridine, an antimetabolite effective in inhibiting *de novo* pyrimidine biosynthesis and treating psoriasis, uniformly is linked to reduced serum zinc concentration and increased urinary zinc excretion.[1495] Administration of 6-azauridine has been associated with the appearances of histidinuria, homocystinuria, and excessive levels

of histidine, cysteine, and homocysteine in blood.[1496]

Other drugs may be associated with total body zinc loss and the appearance of the most common symptoms of it. Drugs that interfere with protein synthesis most commonly produce zinc loss, and they are used in the treatment of malignant processes.[1092] Common clinical symptoms of zinc deficiency observed have been anorexia and weight loss, hypogeusia, hyposmia, dysgeusia, and dysosmia.[671]

However, zinc deficiency in the rat has been associated with a reduction in the metabolic rates of pentobarbital, aminopyrine, and p-nitrobenzoic acid. Microsomal cytochrome P-450 was decreased in livers of these zinc-deficient rats. All biochemical and pharmacologic abnormalities reverted to normal after 14 days of zinc repletion.

Renal Disease and Chronic Dialysis

In patients with proteinuria related to any renal lesion, associated zincuria and a subsequent loss of total body zinc are reflected in decreased serum zinc concentrations. In renal diseases in which proteinuria is not a significant factor, total body zinc levels are variable. In one study of patients with uremia, plasma zinc levels were lower than normal, urinary zinc excretion was within normal limits, and levels of zinc were normal in most tissues except for those in the kidney, which were lower.[308]

The zinc within various body pools of these patients has been redistributed.[308] This relationship has been well documented.[613,975,1440a,1802] In rats, plasma zinc levels decreased after ureteral ligation, but kidney zinc increased.[308] Other organs showed little or no change.

Several changes have occurred in patients undergoing chronic renal dialysis. In early trials with extracorporeal dialysis, loss of total body copper and a subsequent neuropathy occurred. Some attempts to counteract the loss produced acute copper toxicosis.[127,971,975,976,989a] Changes in zinc in blood and tissues also occurred during these episodes, perhaps reflecting zinc content of the dialysis fluids and the tubing as well.[127,1075b] When these materials were purified, many of the acute changes in copper and zinc metabolism were obviated. The values obtained appear to depend, in part, on the type of membrane coils used, but the state of the patient before dialysis and the length of the dialysis may also have their effect.[1816d] Patients undergoing chronic extracorporeal dialysis (with current technology) have a slight but consistent decrease in serum zinc concentration from the zinc found in the dialysate. Although zinc is reduced, a small but consistent increase in copper is found in the serum and in some tissues of dialysis patients (unpublished observations, R. I. Henkin and C. W. Mueller).

It also has been suggested that the way the kidney handles zinc differs from the way in which it handles sodium, calcium, and magnesium.[1547a]

Schizophrenia and Emotional Disorders

Serum zinc levels of schizophrenics have been reported to be lower than in controls;[840,1239] brain autopsy specimens from schizophrenics were found to contain approximately half as much zinc as brains of patients with diseases such as progressive paralysis, congenital syphilis, epilepsy, and erysipelas.[840] The urinary zinc level was elevated in a patient with porphyria and symptoms of schizophrenia.[378a] In this patient, the authors related the changes in zinc levels to the psychiatric symptoms observed, but it is well known that patients with acute porphyria exhibit hyperzincemia. However, in a study of 20 untreated patients with acute and chronic schizophrenia, Gillin (unpublished observations) could ascertain no difference between blood, urine, cerebrospinal fluid, or gastric fluid zinc levels of patients with untreated schizophrenia and controls. These findings are set forth in Table 10-1.

TABLE 10-1

Comparison of Zinc Content in Various Tissues of Patients with Untreated Schizophrenia and Normal Controls[a]

Subjects	No.	Serum, µg/dl	Urine, µg/24 h	CSF,[b] µg/dl	Gastric Fluid, µg/dl
Schizophrenics	20	92 ± 3[c]	286 ± 35	3 ± 1	42 ± 4
Controls	82	92 ± 2	353 ± 23	7 ± 2	40 ± 5

[a] Data from J. C. Gillin

[b] Cerebrospinal fluid

[c] ± 1 SEM

During insulin shock, serum zinc concentrations were reported to increase;[840] it was suggested that the enzymes responsible for glycolysis were zinc-containing enzymes and that insulin treatment increased the activity of carbonic anhydrase. Zinc loss has been correlated with abnormal mental behavior. Zinc-deficient rats were reported to exhibit impaired maze learning compared to pair-fed and ad libitum controls.[950] Zinc-deficient children were reported to perform below normal on cognitive and perceptual motor tests.[1246a] Disturbances of mood have been observed in patients with acrodermatitis enteropathica and kwashiorkor,[1085a] and the acute loss of zinc following L-histidine administration commonly is associated with psychotic ideations, including paranoid delusions, depression, and other profound changes in mental state, symptoms which have been reversed after zinc sulfate therapy.[679]

Other Disorders

Reduced plasma zinc levels were found in patients with psoriasis, various dermatoses, and venous leg ulceration.[559,874] Withers et al.[1794] examined patients with chronic venous ulceration and observed lowered plasma zinc.

Wolff[1802] measured serum zinc in many patients and found it to be lower than normal in many acute and chronic conditions, including hypothyroidism (higher than normal levels were found in hyperthyroidism and hypertension). Lower than normal levels of serum and plasma zinc have also been reported in patients with rheumatoid arthritis.[1252]

Parenteral Nutrition

Fluids rich in amino acids are being given parenterally in increasingly common fashion to assist the alimentation of patients with various disorders when their own absorptive or appetitive mechanisms prove faulty. Parenteral hyperalimentation usually is accomplished with a mixture of essential amino acids, glucose, and electrolytes. Administration of these fluids is commonly followed by losses of

total body zinc,[562,615,1086] perhaps because of the interaction of those amino acids / with the zinc bound to albumin. Albumin-bound zinc is shifted to the amino acids and this amino acid-bound zinc freely passes the renal glomerulus and is excreted in the urine. The loss of total body zinc may become profound and can contribute to the patient's already severe systemic abnormalities. One way to obviate this loss is through the appropriate and judicial use of zinc added to the parenteral fluids administered.

Cardiovascular Disease

Various abnormalities of zinc metabolism have been observed in patients with myocardial infractions. Changes occur in hematic zinc levels[613,632a,999a] and metalloenzymes,[1711] and abnormalities of acute myocardial injury in man[999a] and experimental animals.[632a] It is unclear whether the changes observed in blood zinc levels reflect specific changes related to myocardial function or general changes from stress associated with it or other similar conditions.[462,479a,1466a]

A relationship between decreased incidence of cardiovascular mortality in areas supplied by hard water, has been proposed, although mean serum cholesterol and triglyceride levels were higher in subjects drinking hard water than in those drinking soft water.[111a,333b,873a,982a,1446a] What characteristics of hard water that appear to convey this protection is not clear, but the magnesium and calcium content of the water may be important,[111a] as might be the trace metal content.[982a]

Klevay has suggested that an imbalance between zinc and copper is an important factor in the production of hypercholesterolemia in rats[858] and perhaps in humans.[854a] He has suggested that animal fat ingestion and the zinc: copper ratio in milk may be important in the etiology of cardiovascular disorders in man.[858b] These hypotheses have yet to be fully tested and are not generally accepted.

Vitamin Metabolism

In 1939, Patek and Haig observed that some patients with hepatic cirrhosis exhibited a night blindness that did not improve with treatment with vitamin A.[1213b] Later studies also suggested a clinical correlation between some forms of liver disease and night blindness.[612a,851a] During the acute phase of viral hepatitis, patients not only had serum zinc levels lower than normal,[683] but serum vitamin A and retinol (vitamin A alcohol) binding protein levels were similarly low.[682a]

Although hepatic levels of vitamin A are normal in zinc-deficient rats, hematic vitamin A levels appear to be low,[1512] as are serum levels of retinol and retinol-binding protein.[1514] Concentrations of serum retinol were shown to be lower in zinc-deficient than in zinc-supplemented lambs.[1413]

These studies suggest a relationship between zinc metabolism and vitamin A metabolism. Some of the animal studies were carried out with diets in which less than adequate amounts of protein were fed to the experimental group and pair-fed controls were not systematically employed. Nevertheless, zinc might be necessary to convert retinol to retinene (vitamin A aldehyde)

an energy-requiring process involving the zinc-containing enzyme retinene reductase. This chemical conversion in the retina requires retinene reductase, an enzyme similar to alkaline phosphatase in molecular weight, amino composition, and dependence upon zinc.

Relationships between zinc and the metabolism of several B vitamins,[420] and between zinc and thiamine,[728a] and with pyridoxine,[528] biotin,[282] and folic acid[1781] have been noted. The action of zinc and vitamins on food intake in animals was noted in 1934.[108a]

Feeding either biotin or folic acid had little effect on the growth, food intake, or depleted levels of zinc in liver, hair or serum of growing, zinc-deficient rats.[1201]

Congenital Malformations

There are no direct data which demonstrate a sure teratogenic effect in humans caused by zinc deficiency. However, Sever and Emanuel have deduced from preliminary epidemiologic data that a possible teratogenic effect of zinc deficiency in man may exist.[1475] Some of these speculations were based upon the high rates of malformations in the central nervous systems of infants born in Egypt and Iran, countries where zinc deficiency has been commonly observed.[353a] Hurley[756,757,759,761a] and others[1584,1731,1732] also have speculated that maternal zinc deficiency may cause congenital malformation in humans.[754a]

However, the rat, the model with which Hurley has worked extensively, is highly susceptible to the effects of various teratogenic agents such as antibiotics and cancer chemotheropeutic agents and therefore may not be a relevant model for man. Yet a dam may be fed a zinc-deficient ration

with little or no change in total body zinc content, although the rat fetus may be severely damaged.[756,757] Indeed, the fetus may be damaged even if zinc intake decreased only for a short time during the pregnancy.

Some investigators have speculated that American women may ingest marginal to deficient amounts of zinc.[1404] Thus, it is conceivable that during some periods of pregnancy, particularly in the first trimester, too little zinc reaches the fetus. How this affects fetal growth or development is not fully known. Studies of serum zinc levels generate little useful data because estrogens and other gonadal and placental hormones may depress serum zinc levels regardless of the amount of zinc intake.[1415] Hambidge[616] reported a significant decline in hair zinc in pregnant women between the seventeenth and thirty-seventh weeks of gestation, but the meaning of these reductions cannot be readily evaluated because hair zinc does not always reflect the level of total body zinc accurately with respect to time of sampling.

Porphyria

Abnormalities of divalent transition metals are associated with various types of porphyria.[378a,1171a,1235c,1349a,1360] Patients with active porphyria of several types have been reported to excrete excessive amounts of zinc in their urine,[1235c,1360] although their serum levels of zinc may not be altered.[925a,1360] In 1929, elevated urinary zinc excretion was observed in a patient during an acute exacerbation of porphyria.[378a] Zinc-uroporphyrin chelates in urine have been

measured,[1349a,1738a] and found to be elevated in porphyria patients during acute attacks. Abnormalities of zinc and/or copper metabolism have occurred in patients with the cutanea tarda hereditaria variety.[1235c,1360] These studies led to the use of chelating agents like ethylenediaminetetraacetic acid (EDTA) and British antilewisite (BAL) as antidotes for these conditions. Although these therapeutic regimens were claimed to be successful, data relating porphyria and its underlying abnormalities to trace metal metabolism were not clear enough to establish how trace metals may affect this disease.[1171a] Indeed, conflicting data were presented, and whether abnormalities of metal metabolism are involved in the etiology of porphyria at all is still being disputed.

Levine et al.[925a] studied the zinc metabolism of patients with several types of porphyria. They found that after histidine was administered, the half-time of orally administered zinc-65 shortened significantly, which demonstrated a total body loss of the metal. They also found that during histidine administration, urinary excretion of various porphyrins decreased, although urinary zinc excretion increased.[668,679] This previously observed decreased in excreted porphyrin[1235c] was related by Levine et al.[925a] to the specific action of histidine on the zinc requirement[2,265] of δ-ALA synthetase, the critical enzyme controlling the rate of porphyrin synthesis. Histidine administration dramatically lowered the urinary porphyrin excretion to or toward normal in each patient studied. The role of histidine was found to be useful in reducing urinary porphyrin excretion in these patients.[925a] Because zinc is a component of δ-aminolevulinic acid dehydratase in several species,[2,265] it is possible that abnormalities of zinc metabolism may influence the synthesis of porphyrins.

THERAPEUTIC USES OF ZINC

Wound Healing

Zinc plays an important role in protein synthesis.[515,1410a,1584] It is essential for the normal activity of DNA polymerase and is present in RNA polymerase.[1462] Zinc also appears to influence the synthesis of collagen.[729] Removing zinc from DNA polymerase inhibits polymerase activity, whereas complete activity is restored after its incubation with zinc ions.

Physiologically, zinc is important to growth and development. Some bacteria require zinc to synthesize several essential amino acids.[108] Zinc is also an essential nutrient for higher plants.[1523] Animals on zinc-deficient diets develop anorexia, hypogeusia, growth retardation, and various skin dystrophies.[270,271,1004] Each sign of zinc deficiency found in animals has also been reported in man, although the relationship between zinc deficiency and the abnormalities observed have often been difficult to specify.

These biochemical and physiologic observations indicate the importance of zinc to cell division in several systems of different species. Therefore, it is not surprising that zinc would affect any system undergoing rapid cellular division, such as a healing wound. Such an observation was made in apparently healthy men undergoing surgical excision of pilonidal cysts.[1262,1263,1265] In this study, conducted without the benefit of systematic controls, orally administered zinc was claimed to accelerate the healing rate of surgical wounds.

Zinc may accelerate healing because zinc pools shift radically after an operation. In the postoperative or wounded period, more zinc is necessary to promote protein synthesis, collagen formation, or the incorporation of zinc into enzyme systems. Radioactive zinc has been found to concentrate in healing tissue, and highest levels have been found immediately after injury.[1418] The greatest activity of zinc in wound healing has been suggested to occur during epithelization when the large store of zinc in the skin may be a convenient source of the metal.[1265] Zinc has been shown to preferentially concentrate in healing tissues, with peak levels reached the third day after injury.[1418]

Serum and urine zinc have been reported to be lower than normal in patients with bedsores[1] and interpreted to reflect the depleted body zinc of these patients.[295] Oral zinc sulfate has been said to promote healing.

Zinc given to animals to aid wound healing is not consistently effective in increasing the healing rate.[1054,1058,1096,1411] These studies in animals and in man[75a,291b,559a,1102b] cast doubt on claims that zinc is influential in wound healing.

Other controlled and uncontrolled studies[75a,295,504c] have been performed to evaluate the claims that zinc accelerates the healing rate of surgical wounds. Single- and double-blind studies involving oral administration of zinc and placebo have been carried out in man and animals and have produced conflicting results.[291b,559a,762a,1102b,1305b,1471a] Some verified the efficacy of zinc in accelerating the rate of wound healing.[559a,762a, 1265,1305b,1471a] Others did not.[75a,291b,1102b] But if zinc plays such an important role in cell division and protein synthesis, how can the negative results be explained?

These studies are problematic. How can the rate or end point of the healing process be established to the equal satisfaction of different investigators? The absorption of oral zinc by humans is variable-dependent upon several factors, including the counterion used and the nutritional status of the patient; the results of different studies cannot always be compared directly. Evaluation of the results may also be difficult because human zinc absorption is slow and variable during the first few weeks of oral intake. As long as 6-8 wk may be required before some aspects of bodily changes in zinc metabolism can be measured.

Although no definitive answers have been found, a double-blind study was reported in which the effects of zinc and placebo were studied on the rate of healing of venous leg ulcers.[605] If all data from that study were pooled and the total results analyzed, no significant difference would be found between zinc and placebo in accelerating the rate of wound healing. However, when patients with lower concentrations of serum zinc were evaluated as a group, a statistically significant acceleration of the rate of

healing was demonstrated, whereas no differences could be shown in patients with what investigators considered normal concentrations of serum zinc. Similar outcomes (that is, open to two interpretations) were also demonstrated in zinc-deficient rats and rats on diets supplemented with zinc.[1411] These results suggest that zinc aids wound healing in zinc-deficient states, but not in states where zinc is adequate. If these latter studies are confirmed, the only problem would be to identify

the patient or subject group at risk.

Unfortunately, this latter task is simpler to state than to solve. In rats on a zinc-deficient diet, the serum zinc concentration may be an adequate index of zinc deficiency.[1522] However, in humans, serum zinc concentration may not accurately reflect the status of total body zinc metabolism. Acute or chronic infectious processes, anemias, various drugs, liver diseases, malignancy and other pathologic processes may depress the concentration of serum zinc and affect a redistribution of body zinc pools, yet not necessarily produce a total body zinc loss or a state of zinc deficiency. How then is the patient at risk to be readily identified?

No simple answer exists although several approaches are possible. The clinical ideal would be a simple, direct method of assessing total body zinc status with one quick and easy test. Since assessing status of the serum zinc pool may be misleading, measurement of urinary zinc excretion along with it may be helpful. However, this index also must be used cautiously, for during decreased food intake or starvation human serum zinc concentration may be normal, whereas urinary zinc excretion may be quite elevated. Any bodily process which involves the breakdown or rapid turnover of cells is associated with increased urinary zinc excretion,[1532,1537] changes which may or may not be reflected in serum zinc concentration. Since mobilization of the hypothalamic-pituitary-adrenal axis is associated with decreased serum zinc concentration and increased urinary zinc excretion, stress from any significant source, including surgery itself, could be manifested by a redistribution of body zinc and a transient zinc loss, both of which may be evident for a few days. Measurement of hair zinc is relatively simple but it does not reflect

the dynamic changes inherent in body zinc metabolism. Measurement of salivary zinc[678] may offer a direct index of total body zinc, but too little is known about this new technique.

Administering zinc to zinc-deficient humans or animals may well correct the numerous defects produced by the deficiency. The mechanisms by which these abnormalities are corrected are multiple, and they are related to the specifics of the several organ systems affected by the deficiency state. However, /the incidence of zinc deficiency in man has not been established clearly, and as in most disease states, /it is represented by a spectrum of severity. Secondary effects of zinc, unrelated to net total body zinc loss, also may be important, particularly in organ systems which metabolize rapidly. If the role of zinc in cell division and wound healing in man is to be comprehended, changes in the intake, absorption, excretion and distribution of zinc, as well as the many biochemical, physiologic and pathologic factors which influence the dynamic nature of these processes will have to be evaluated carefully.

States of Zinc Deficiency

Clinical zinc deficiencies in humans have been related to zinc malabsorption caused by gastrointestinal diseases of several etiologies,[150a,214a,998a,1412,1522] decreased zinc intake, and loss of zinc-rich fluids.[296,899a,899b,899c,1793a] Urinary losses following hepatic and renal damage, the effects of certain drugs, excessive perspiration,[309a,720a,1288,1585a] or some combinations of these phenomena also may be responsible for zinc deficiency. Whatever the etiology, clinical symptoms of anorexia

and taste and smell abnormalities have been considered early signs of insufficient zinc.[673,679]

Taste and Appetite Disorders

The role of zinc in appetite has been studied in normal subjects,[679] and those with scleroderma[679] or malignancies. After zinc was depleted by L-histidine administration, anorexia uniformly developed as the first symptom associated with zinc loss. Appetite returned to normal after oral zinc administration.[673,679] In some patients with malignancy and cachexia, treatment with zinc ion has been useful in obviating some of the more severe effects of anorexia; in others with idiopathic hypogeusia and anorexia, oral administration of zinc ion is useful in obviating anorexia if the hypogeusia has been corrected.[681, 1420b] In the rat, anorexia and a marked cyclic pattern of food intake also developed as the first symptoms of zinc deficiency and these symptoms were the first corrected even with the administration of very small amounts of zinc.[270]

Acute or chronic zinc deficiency or loss in humans[615,617,620a,621,679,1522] and animals[1004] has been accompanied by anorexia, hypogeusia and hyposmia. As noted above, anorexia is the initial symptom in all species. The symptoms of hypogeusia, hyposmia, dysgeusia and dysosmia have been shown to develop later.[678] Oral administration of zinc to human patients with acutely induced zinc deficiency abolished the dysgeusia and dysosmia and restored taste and smell acuity to or toward normal in each patient so affected.[679] Changes in human taste acuity were related to the diminution of the recently isolated parotic zinc protein, gustin, which has a molecular weight of 37,000 daltons and contains 8% histidine and 2 moles of zinc/ mole of protein.[674] This protein is present in subjects with

normal taste acuity but has been shown to be decreased in patients with various types of taste dysfunction.[674] Because this protein contains zinc and is the major zinc-containing protein in saliva,[674] measuring salivary zinc excretion from the parotid gland may be a convenient way to obtain information about the concentration of this protein in parotid saliva.[674,678] Decreased zinc concentration in parotid saliva often reflects decreases in salivary gustin concentration;[674] this change in turn is commonly associated with anatomic abnormalities in the taste buds.[674]

The pore area of the bud is disrupted by several pathologic processes, but they may be observed systematically only with the use of transmission electron microscopy. Pathologic conditions may be reflected by several anatomic and functional abnormalities. In patients who lack saliva, e.g., patients with xerostomia from Sjögren's syndrome or fatty parotid syndrome, there is a decrease in taste acuity (hypogeusia), and in those taste buds that can be found, the pore area of the bud is disrupted.[684] Treatment of these patients with drugs or x-irradiation, in whom salivary flow was restored to or toward normal, was accompanied by the restoration of normal taste bud anatomy and taste acuity to or toward normal.[684] Patients with untreated Sjögren's syndrome and hypogeusia do not exhibit decreased levels of salivary zinc; rather they have normal levels of parotid salivary zinc but apparently too little salivary secretion to support normal taste bud function.

The salivary flow rate of patients with idiopathic hypogeusia (i.e., taste loss related to unknown or unexplained factors) is normal, but taste acuity is impaired.[674,678,684a] Their taste bud anatomy is altered in a manner grossly similar to that observed in patients with xerostomia — the pore area of the taste bud is markedly disrupted and the normal cell types exhibit cytologic pathology. In some patients with idiopathic hypogeusia, analysis of parotid saliva revealed lower than normal levels of zinc and gustin. 674,678

Treatment with zinc ion in a single-blind study restored taste acuity and taste bud anatomy to or toward normal in some patients with idiopathic hypogeusia, and in both single- and double-blind studies, concomitant increases in parotid salivary zinc and parotid gustin concentrations were observed.[673a,674,678,681] For other patients not only was zinc ion ineffective in restoring taste acuity and taste bud anatomy to or toward normal, but also their parotid salivary gustin concentrations did not increase.[673a] Placebo treatment was also effective for some patients, as noted later, and ineffective for others in restoring taste acuity to or toward normal as observed in a double-blind study.[673a] No systematic studies have yet been published in which the nature of metabolic or other changes in these patients improved by placebo have been ascertained.

The rationale for treating patients with idiopathic hypogeusia with zinc ion lay in empirical observations of taste and smell dysfunction and abnormalities of zinc metabolism among patients with acute viral hepatitis,[682a,683] and in women during the first trimester of their pregnancies. In both of these groups, taste and smell dysfunction returned to or toward normal when zinc metabolism returned to or toward normal with the waning of the hepatitis or the progression and subsequent termination of the pregnancy. In a single-blind study of the influence of zinc ion and placebo in treating patients with taste dysfunction of several etiologies, a statistically significant effect of zinc ion was measured.[681] In addition, hypozincemia was observed among these patients, reinforcing the hypothesis of an abnormal zinc metabolism. In a subsequent double-blind study[673a] in a similar group of patients, both placebo and zinc ion were equivalent in restoring taste acuity to or toward normal. Reevaluation of the double-blind study, carried out in the same manner as the single-blind study, suggested that the order in which the drugs were given affected the results of the study.[673a] Thus, zinc therapy is clearly not effective in the treatment of unselected patients with taste disorders.

Although taste dysfunction has been described as a common accompaniment of zinc deficiency states and treatment with zinc ion has been shown to correct the abnormalities of the deficiency, including those of taste and smell dysfunction, patients with changes in taste and smell function do not always exhibit abnormalities of zinc metabolism. As noted, available data indicate that placebo therapy cannot be differentiated from therapy with zinc in the correction of taste and smell dysfunction of diverse etiologies.

The confusion between zinc deficiency in which taste loss is a symptom, and taste loss, which may be related to a variety of factors, is compounded by the lower than normal mean serum levels of zinc observed in patients with taste and smell dysfunction when measured as a group.[673a,681,1420b] Before the double-blind study carried out by Henkin and his associates,[673a] it was assumed that most patients with taste and smell dysfunction exhibited an abnormality of body zinc metabolism because of their low serum values. This hypothesis was buttressed by the finding of decreased alkaline phosphatase in leukocytes of patients with hypogeusia participating in the double-blind study.

However, these findings may not reflect total body zinc loss. In a study of the oral absorption of zinc-65 in 13 patients with hypogeusia and hyposmia, three groups of patients were tentatively identified.[1a,950a] Six patients absorbed 65 $\pm$ 4%(mean $\pm$ 1 SEM) of the zinc-65, a value similar to that reported[950a,1345c] for normal subjects. Five patients absorbed 22 $\pm$ 5% of the dose given, a value similar to that reported as indicative of zinc malabsorption. This value is similar to that absorbed by patients with untreated acrodermatitis enteropathica. Two patients

absorbed 99 ± 1% of the administered dose. Athough the mean level of serum zinc in all 13 patients was lower than normal, only 30% exhibited apparent "zinc malabsorption."

The role of zinc in the taste system has been investigated in several studies. Taste bud-enriched membranes have been isolated from bovine circumvallate papillae.[958d] As these membranes were purified, the activity of alkaline phosphatase, a zinc-dependent enzyme, was significantly enhanced and found to be the major enzyme of the purified membranes.[958b] In addition, the specific binding of several tastants to these purified membranes was inhibited by EDTA but rose toward normal by adding zinc ion.[958b] The saliva and mucus secretions from the small gut of zinc-deficient sheep and rats are also different from their normal counterparts[1307a] and these secretions return to normal when zinc ion is provided.

Color Blindness

Michaelson noted that in patients with acrodermatitis enteropathica whose color blindness was induced from optic atrophy after diiodohydroxquin treatment, zinc restored color vision to normal.[1037b] The visual cones may be sensitive to zinc deficiency and thus zinc might influence the visual process.

Treatment of Laryngeal Granulomas

Granulomas of the larynx arise from several etiologies and often are difficult to manage. Since repeated surgical removal may be complicated by repeated regrowth of this tissue, narrowing of the laryngeal airway, and other surgical problems, a chemical method of treatment has been sought. Oral administration of zinc has been reported to be

useful therapy.[1305, 1305a, 1305b] The specific relationship between zinc therapy and the occasional complete disappearance of these lesions is not clear at present, although the rationale for its use is based on the hypothesis that zinc accelerates the healing of wounds.[1418]

Skin and Epithelial Tissue

Locally applied and orally ingested zinc has been used to treat several skin disorders. Topical application of calamine, an impure zinc oxide, was described as early as 1550 BC. Calamine is still widely used as a proprietary preparation to treat many skin disorders and zinc oxide itself has been prescribed by dermatologists for many years. Zinc is chief constituent of many common proprietary agents, including talcum powders and skin creams. Zinc stearate, oleate, and sulfate solutions are main ingredients in talcums and creams. Zinc has also been claimed to rejuvenate hair and skin.[296] Dilute solutions of zinc sulfate and acetate are components of eye washes prescribed to treat conjunctivitis; and they are sometimes applied as ancillary aids in the management of urethritis. Zinc peroxide pastes are commonly used in the treatment of skin ulcers, especially sores that are complications of varicose veins or vascular insufficiency of the lower extremities.

Cardiovascular Disorders

Abnormalities of zinc metabolism in several disorders, including hypertension, arteriosclerosis, and Raynaud's phenomenon have been directly or indirectly related to the pathogeneses of those disorders.[685b,686] Treatment with zinc ion has been used to reverse the abnormalities observed in atherosclerotic cardiovascular disease.[686a, 1261,1557a] The validity of these impressions remain to be clearly established, because clinical trials have not been controlled. It has been / claimed that zinc improves local blood flow to ischemic

areas in patients with impaired circulation to the leg;[686] this hypothesis has not been rigorously tested yet. However, zinc-65 appears to deposit preferentially in rapidly proliferating tissue, particularly areas undergoing rapid healing.[685a,843a]

Routes of Zinc Administration*

In general, when zinc is administered to animals or humans, it is given by the oral route.[1262,1263,1265] Effectiveness of oral absorption depends upon the many factors noted in Chapters 7 and 8. At present, sulfate is the only approved, easily available counterion for pharmacologic zinc therapy in the United States.[1647] Nevertheless, this compound is poorly absorbed and one of the more toxic of the available zinc compounds.

Oral administration of zinc sulfate may be associated with gastrointestinal toxicosis in some subjects. The Food and Drug Administration has approved the use of zinc only as an emetic for humans. Thus, administration of significant amounts of oral zinc is limited by this side effect.

The pharmacology of oral zinc administration and subsequent blood and tissue levels have not been studied systematically. Similarly, pulmonary inhalation of zinc and subsequent changes in blood and tissue levels have not been carefully investigated. Since zinc is used to treat diverse disorders, its pharmacology is of immediate and practical interest to physicians and veterinarians.

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* Because of the limited state of knowledge and the preference for oral administration, intravenous and other kinds of parenteral administration of zinc will not be discussed. However, these routes have become more important in treating some clinical conditions, particularly in patients undergoing long-term hyperalimentation.[615]

CHAPTER 11

TOXICITY OF ZINC

HUMANS

Ingestion, Inhalation, and Absorption

Acute effects. Apart from effects of inhaling zinc fumes encountered during some types of industrial exposure, it is apparent that zinc, especially from oral ingestion, is not very toxic to humans. There are a few sparse reports of accidental exposure of humans to food or drink contaminated with high levels of zinc. However, these reports are problematic because it is difficult to determine whether the alleged zinc toxicosis was caused by the zinc *per se* or by some impurity associated with the metal, particularly cadmium. In addition, the literature is old and lacks present day confirmation.

Acute onset of gastrointestinal distress and diarrhea was reported to follow ingestion of lemonade prepared in galvanized iron garbage cans for military personnel stationed in Panama.[222] Zinc absorbed into the acidic beverage was suspected of being responsible for this outbreak of diarrhea, and use of galvanized iron utensils for preparing or serving food or drink was subsequently prohibited by the Army Medical Research Board. Another incident was reported in which several people became ill, apparently with food poisoning, from food eaten which had been cooked and/or held in galvanized containers.[173] Storing food, particularly acid foods such as citrus punches, in galvanized containers may bring about considerable zinc contamination.

An even less well documented report was a letter which described a couple who drank water containing 40 ppm zinc from a galvanized pipe.[900] They became irritable, and suffered difficulty in concentrating, drowsiness, cloudiness of thinking, mental fatigue, and headaches. A diagnosis of zinc toxicosis was only suggested by the nonspecific symptomatology and the couple was said to have improved upon change in water supply.

More recent reports suggest that ingested zinc is toxic to humans only in very high dose levels that break down the homeostatic mechanisms controlling zinc uptake and excretion.[124] Eighteen patients were given a daily dose of 660 mg zinc sulfate for 16-26 wk for treatment of venous leg ulcerations. No evidence of hematologic, hepatic, or renal toxicosis from zinc was found in any of these patients.[560] It was reported that a 16-yr-old boy ingested 12 g of elemental zinc in 2 days to hasten healing of a minor laceration. In 3 days he developed a lethargy that progressed for 5 more days. He complained of light-headedness, and exhibited a slightly staggering gait and difficulty in writing. These effects were accompanied by an elevated blood zinc concentration and increased serum amylase and lipase, suggesting a pancreatic effect.[1094]

A patient on home hemodialysis experienced nausea, vomiting and fever, allegedly from zinc toxicosis that developed when the metal contaminated the water stored in the galvanized tank of her dialysis unit. Plasma and red blood cell zinc concentrations were elevated.[520]

Chronic effects. Whether long term or chronic ingestion of excessive zinc in humans is associated with adverse health effects is debatable. A syndrome consisting of gastrointestinal symptoms which include anorexia, nausea, vomiting, epigastric discomfort, and weight loss was described in workmen in a galvanizing plant as characteristic of excessive exposure of zinc.[1006] However, simultaneous exposure to other potentially toxic substances also occurred, so that identification of zinc as the principal toxic substance is uncertain.

Another report suggests occupational exposure to zinc oxide fumes at concentrations ranging from 3-15 mg/m^3 for 2-35 yr may not affect health adversely.[84] Increases in blood zinc and zinc content of gastric secretions were found in furnace operators who were chronically exposed to zinc oxide fumes in a brass foundry in Egypt.[622] The increased zinc concentration in gastric secretion might account for the reported gastric complaints, notably epigastric pain after food intake. Radiography detected no pathologic changes in stomach or duodenum in the four workers studied by this technique.

On the basis of a single case report, it has been suggested that chronic inhalation of zinc stearate powder may produce chronic pneumoconiosis.[1145]

Some observations of children have suggested that excessive oral intake of zinc might be more widespread than previously suspected. For example, toy cars made with zinc are often placed in warm bath water along with soap. Because of the high pH, this environment would be conducive for high concentrations of zinc to leach out of the toy into the bath water. Children do drink bath water, and this chain of events has been reported to precipitate the lethargy, fatigue and acute hemolytic anemia of some children observed in hospital.[284] Their serum zinc concentrations were elevated, as was the amount of zinc excreted in their urine.

Excessive intake of zinc in the form of zinc supplements given to aid healing of wounds has produced lethargy in children, along with high levels of blood zinc.[1094] A severe porotic effect of oral zinc on bone also may result, especially if dietary calcium is low.[463] Other investigators have noted that oral zinc supplementation is not without medical consequences.[251]

Metal Fume Fever

Metal fume fever is defined as an acute disability of short duration that occurs when fume is inhaled from metal heated to a temperature above its melting point.[751] Clinical features as well as the pathogenesis of the syndrome have been summarized.[177,751,829,1555] The disorder has been most commonly associated with inhalation of zinc oxide fume, but it may be seen after fumes of other metals are inhaled, particularly magnesium, iron, and copper. But metal fume fever is most severe among brass founders and the higher the proportion of zinc, the more severe are the symptoms.

Clinical signs and symptoms occur within 4-8 h of exposure. They are characterized by hyperpnea, shivering with fever, profuse sweating, pain in the chest and legs, and general weakness. An attack is usually short--lasting only 24-48 h--and accompanied by leukocytosis. One of the more intesting aspects of this disorder is that a second exposure to the metallic fumes within 48 h will produce little or no response, but if the second exposure comes after a lapse of 48 h or more, an attack is likely. This is best demonstrated in workmen who suffer fresh attacks on a Monday following a weekend away from exposure. However, if the individual continues to be exposed daily, he does not become ill again. The word "tachyphylaxis," meaning "quick immunity," has been applied to this phenomenon.[1005]

Although metal fume fever was said to be recognized as long ago as 1822 by the French physician, Potissier,[751] complete understanding of the role of zinc in the pathogenesis of the syndrome is still not available. McCord[1005] refers to metal fume fever as the foremost scientific enigma

of industrial toxicology. Several theories have been advanced to explain the observed features of the disorder but few of these notions have received experimental support.

One appealing hypothesis is that inhaled zinc fume has a direct chemical toxicity to alveolar and capillary epithelium, which brings about an acute inflammatory response and consequent exudation.[177] Fume fever, the systemic reaction, may occur because protein produced by breakdown of polymorphonuclear neutrophils pours into the circulation. This concept is a slight variation of an earlier proposal[919] that the inhaled zinc produces pulmonary bacteriolysis. Proteins released from the lysed bacteria are pyrogenic. A more recent study suggests that protein released from cell breakdown serves as an endogenous pyrogen.[1230] Extracts prepared from tracheal mucosa and lungs of animals with experimentally induced metal fume fever have produced similar symptoms when injected into other animals.[1631]

McCord[1005] suggested that the effects of zinc on respiratory tract tissues result in the formation of an allergen and eventually to an antigen-antibody, or hypersensitivity reaction. The thesis is intriguing but it has not been proved. Also, one might expect eosinophilic polymorphonuclear leukocytes to increase significantly as part of the peripheral leukocytosis occurring in the disorder, but apparently no such process occurs.

Another experimental approach has been to reproduce the disorder in animals by spraying blood serum into a zinc oxide cloud, and collecting and injecting this material into animals.[1430] It also was claimed that the syndrome could be induced by the subcutaneous or intravenous administration of zinc salts,[207] but this method has not been repeated or verified. In fact, there is little or no evidence to suggest that metal fume fever is a direct result of systemically absorbed zinc per se.

Some of the most detailed clinical and experimental studies of the pathogenesis of metal fume fever here conducted by Drinker and his associates between 30 and 40 years ago.[398,399,402] It was demonstrated that an initial respiratory exposure to zinc fume at 52 mg zinc/m^3 air was capable of inducing metal fume fever in human subjects, although later exposure to higher concentrations within 24 h had no further effect. They believed then and it was more recently contended by some[1005,1358] that freshly formed fumes composed of very small particles from 0.05-0.5 μm are necessary to cause the disorder. As fumes age, they tend to agglomerate and become less reactive. Larger particles tend to settle out or become entrapped higher in the respiratory tract.[402]

Drinker and Drinker[399] also produced clinical metal fume fever in cats, rats and rabbits by exposing them to high concentrations (600 mg zinc/m^3 air) of zinc oxide fumes. Within 1 or 2 h the animals exhibited an initial decrease in body temperature followed by fever. They also demonstrated that the highest concentrations of zinc in these animals after pulmonary exposure were in pancreas, liver and gallbladder, a phenomenon similar to that found after the feeding of a single dose (0.175-1.0 g) of zinc oxide. They also learned that elevated atmospheric carbon dioxide, perhaps because it increased depth of respiration, increased the severity of experimentally induced metal fume fever.

It has been shown that the inhalation toxicity of catalytically active zinc oxide dust is increased markedly by ultraviolet radiation.[94]

Krause[876] demonstrated that inhaling zinc inhibited red blood cell carbonic anhydrase activity; the level of inhibition correlated with dose of zinc. Similar inhibition of carbonic anhydrase followed inhalation of

magnesium oxide, a metal that also produces metal fume fever. He suggested that inhibition of carbonic anhydrase was the actual cause of metal fume fever, but beyond demonstration of the association, the rationale for this notion was not discussed.

Reasons for the transient tolerance or immunity of experimental animals and exposed workers to metal fume fever also remains unexplained. Drinker[402] observed that the period of immunity or tolerance seems to parallel the period of the leukocytosis.

It also must be considered whether or not the symptoms of metal fume fever that seem to be caused by the metallurgy of zinc are actually arising from the presence of a metal contaminant, such as arsenic, cadmium, manganese, lead, chromium, or silver. Apart from the knowledge that the syndrome may be contracted from exposure to other forms of metal fume, particularly manganese oxide, there is no other information implicating these metals.[697] Moreover, a similar syndrome (cotton-mill fever, or byssinosis) occurs among cotton mill workers,[1585] presumably from exposure to a foreign protein in cotton dust.[356]

Therefore, it can be said that exposure to finely divided zinc dust will produce a disorder described as metal fume fever. The pathologic mechanisms involved in the expression of the syndrome are not yet defined. The syndrome may be produced by other forms of metal fume or protein-containing dusts.

ANIMALS

Animals are quite tolerant of high levels of zinc in the diet. Levels 100 times that required in the diet usually do not cause any detectable

symptoms of toxicosis. Part of any observed toxicosis is caused by the decreased food consumption that accompanies the feeding of high zinc diets; interference with copper and iron absorption and metabolism is probably also responsible for toxicosis.[1640] High zinc ingestion frequently affects the joints of animals fed such a diet; this effect is probably not from interference with food intake or copper and iron metabolism, and may be related to the increased level of zinc in the bones of animals fed excess zinc. The adverse effects of high levels of dietary zinc on bone have been reported to be aggravated in the presence of low calcium intake.[464] Levels of zinc as high as 600 ppm in the diet have not been reflected in hair.[1051,1198] Elevated plasma zinc coupled with low plasma copper has been suggested as a reliable indicator of chronic zinc intoxication.[1062a]

Horse

Reports of lameness and death in young horses raised in the vicinity of lead-zinc smelters prompted a study of lead and zinc toxicosis in young horses.[1786] Growth rates of animals fed zinc decreased after intake exceeded 90 mg/kg body weight/day. Enlargement at the epiphyseal region of the long bones was the first clinical sign noted, and it occurred in all animals between 20-30 wk. These swellings were followed by stiffness, reluctance to walk and turn, lameness, and reluctance to stand. The growth of animals receiving lead was similar to that of controls until signs of illness appeared. Animals that received both lead and zinc had lower levels of lead in bones but higher levels in liver and kidney than did those fed only lead. They did not,

however, have the neurologic dysfunction associated with the high levels of lead.

Another report of illness in foals in the vicinity of a smelter was diagnosed as lead toxicosis,[1431] but the symptoms are more like those described for the horses given either high levels of zinc or of zinc and lead. Since zinc in grasses was as high as 1,100 ppm and 3,500 ppm in overwintered grass, it is likely that the condition resulted from the high levels of zinc as well as lead. Since a foal affected the summer before the study was undertaken was still stiff the next summer, it would appear that horses do not recover once they have been affected, although here the animal was still eating the contaminated forage. Older horses and cattle in the area were not affected.

A German study in which similar involvement of bone joints was found in foals pasturing near metalworks with large lead and zinc emissions indicated that cattle were also affected at times, although not as severely.[754] If not too severe, damage to the joint was apparently reversible; but both horses and cattle were likely to suffer other complications which decreased their usefulness for breeding.

Swine

A zinc-lead interaction has also been demonstrated in growing pigs. Zinc (0.4% of the diet) increased lead (0.1% of the diet) toxicosis when fed over 15 wk.[728] The toxicity was greater with low levels of calcium-phosphorus in the diet.[734] (Zinc interferes with development and mineralization of bone by decreasing calcium and phosphorus content as well as reducing the calcium:phosphorus ratio.[1391]) Enlargement and

softening of epiphyseal ends of the humerus and femur were reported in young pigs receiving milk piped through galvanized iron piping and attributed to high levels of zinc in the milk.[568] In swine overdosed with dietary zinc (0.4%), zinc levels in the liver increased, whereas hepatic iron decreased markedly and copper remained unchanged.[328] Pigs fed levels of zinc alone up to 500 or 1,000 ppm for several weeks showed no significant difference in weight gain.[61,1640]

Ruminants

Calves fed 600 ppm zinc for 7-21 days showed no sign of toxicosis although zinc levels in pancreas, liver, and kidney were strikingly elevated.[1051,1060,1544] Lactating dairy cows receiving 1,300 ppm zinc for 6 wk did not have a greater increase of zinc in their milk than cows on a 700 ppm zinc diet. Moreover, feed containing 1,300 ppm did not produce any discernible changes in health or milk production.[1050] Levels around 1,000 ppm appear to be approaching toxic levels for lambs and feeder cattle, however.[1640] Levels of this order have been found in mixed pasture herbage downwind of possible zinc sources.[1063] Grasses grown in an area in which a smelter previously had operated had levels of 600 ppm.[890] The greater toxicity to ruminants may be accountable to adverse effects on rumen microorganisms, since cellulose digestion by rumen bacteria *in vitro* was reduced by zinc concentrations of 10-20 μg/ml.[825,981] The effect varied to some extent with the protein source.[825]

Cows that accidentally ingested a very large dose (20,000 ppm) of zinc oxide contracted severe enteritis and prostration, and some fatalities followed.[20]

Poultry

Mallard ducks fed high levels of zinc suffered severe paralysis; some ducks were unable to walk after 30 days.[524] The lowest level fed them was 3,000 ppm, but even at this level food consumption decreased. At higher levels consumption was reduced to the point that none of the groups received as much zinc in a 30-day period as did those on the 3,000 ppm intake. The high mortality in the groups receiving over 3,000 ppm zinc may have been caused primarily by decreased feed intake.

Addition of 3,000 ppm zinc to a corn-soybean meal ration significantly reduced growth in chickens, too. Only a slight reduction occurred in growth with 2,000 ppm,[796,1351] and taking chicks off the high zinc diets brought about weight gains during the next 6 wk equal to that of the controls.[796] When given in a sucrose-fishmeal diet, 2,000 ppm zinc did impair growth, but this diet did not support normal growth even with control levels of zinc.[96] Mortality from Salmonella gallinarum was not affected by 2,000 ppm zinc in the diet.[702] Growth in turkey poults was reduced slightly by 4,000 ppm zinc but not by 2,000 ppm.[1700]

Cat and Dog

Cats fed daily doses of zinc oxide between 150-600 mg and dogs given 500-1,000 mg daily for periods from 3 wk to 1 yr showed no sign of damage.[400] Although in this experiment cats did not reject the food until the zinc oxide concentration was greater than 600 mg, other investigators found that cats either rejected or vomited a diet containing more than 300 mg zinc oxide.[1458] After 12-16 wk on a diet containing 300 mg zinc oxide, cats lost weight and exhibited marked fibrotic changes in the pancreas as well as a decrease in pancreas size.[1458]

Dogs given 4 mg zinc gluconate/kg body weight intravenously developed lassitude, enteritis, and paresis of the hind legs.[1652] Their electrocardiograms recorded changes similar to those charted in cases of potassium intoxication.

Rat and Rabbit

The rat can tolerate very high levels of dietary zinc; the median lethal dose of ingested zinc for rats is 350 mg/kg body weight.[223] A level of 2,000 ppm in the food during gestation and lactation had little effect on either dams or offspring.[330,1426] Feeding 5,000 ppm during gestation did not affect maternal weight or number of viable offspring, although fetal weights and weights of 14-day-old pups were decreased.[839] If females were fed 5,000 ppm from weaning, however, their litter size was reduced, and most of the offspring were born dead.[1580] Feeding 4,000 ppm for 3 wk before mating also reduced litter size.[1426] In another study, however, females that received 7,000 ppm zinc from weaning were able to maintain pregnancy, although litter size was reduced and only 50% of the fetuses were viable.[457] If 4,000 ppm were fed during lactation, the pups had increased zinc and decreased iron and copper in the liver.[281] To some extent the hepatic condition was a reflection of the higher zinc level (approximately three times normal) and lower copper and iron levels in the milk;[280] but since the pups had access to the dam's diet, the pups' ingestion of the high zinc diet probably was responsible for much of the change in mineral levels.

Consumption of high zinc diets accounted for decreased copper concentration in some tissues, particularly liver. Interference with copper metabolism was reflected in lessened activity of cytochrome oxidase and catalase.[1682] Enzymatic activities returned to normal in both liver and heart[407] if the high zinc diets were supplemented with copper. In young rats

liver copper was reduced on a 2,000 ppm zinc diet more than it had been on a 1,000 ppm diet.[1761] Zinc concentration in liver, kidney, and spleen was much higher with 2,000 ppm zinc in the diet than with 10-1,000 ppm.[266]

Intake of a high zinc:copper ratio (40:1) has been reported to cause hypercholesterolemia in rats.[858] High zinc:copper ratios were therefore suggested to be of concern in human diets because of the association of hypercholesterolemia with heart disease.[854,858a] The hypercholesterolemia in rats, however, may have been caused by copper deficiency because the level of ingested copper was quite low. Since zinc and copper are antagonistic in several aspects of metabolism,[362a] a high zinc:copper ratio may induce copper deficiency when copper levels are low, but have no effect when they are higher. For instance, zinc:copper ratios of 10:1 and 20:1 reduced weight gain in weanling rats at a copper level of 1 ppm but not at 2 ppm.[1099] Even with a low level of copper in the diet, a zinc:copper ratio of approximately 20:1 did not increase serum cholesterol in weanling male rats fed the diet for 4 wk.[442] Supplementing infant formula with a zinc:copper ratio of 17:1 for 6 mo did not increase serum cholesterol.[1722] Zinc sulfate injected subcutaneously reduced serum cholesterol in adult rabbits.[1560] Zinc was given in drinking water (35-50 mg/day) to rabbits on control diets and on atherogenic (cholesterol-supplemented) diets. Rabbits fed the atherogenic diet showed decreased serum zinc levels and, compared to controls, zinc concentration in their aortas was reduced.[472] High levels of zinc in the diet decreased fat content in the liver of young rats on a high fat-low protein diet.[1391]

Cardiovascular lesions have been linked to copper deficiency in the rabbit.[750] In rats, excess zinc decreases cytochrome oxidase activity of the heart, but

copper supplementation will restore cardiac oxidase activity to normal[969] or greater than normal.[407] Similarly, high dietary levels of zinc markedly decrease liver catalase and cytochrome oxidase activities, but they return to normal with addition of copper.[1682] However, copper deficiency in Americans appears to be extremely rare.[1117] Since no evidence exists of marginal copper deficiency in the general population of this country, there is no reason to expect high zinc:copper ratios in the diet to cause hypercholesterolemia.

In pregnant rats and their offspring, high levels of dietary zinc also reduced iron concentrations in some tissues,[281,326,839] but that effect was less marked and more variable[330,839,1426] than the effect on copper. In young rats, 4,000-7,500 ppm zinc produced a condition resembling iron deficiency anemia and reduced hepatic iron stores.[329,1516] Supplements of iron plus copper increased hemoglobin concentration to normal levels, although growth remained depressed.[969] A level of 1,200 ppm zinc in the diet of young rats was reported to cause decreased hematocrit,[742] although weight gains were not affected. It would be wise to check animals on high zinc intakes for signs of anemia or copper deficiency even if growth does not appear to be affected.

Reports of the effects of zinc on the kidney are sparse. One study suggested that rats given intraperitoneal injections of zinc chloride on alternate days for 10 doses of 2.4 and 4.8 mg/kg body weight would develop histologic changes in renal tubular lining cells.[454] The nuclei enlarge, and the formation of intranuclear inclusion bodies is reminiscent of the changes that occur in lead or zinc intoxication. Repetition of this experiment in another laboratory failed to produce those changes (R. A. Goyer, personal communication).

Walters and Roe found no indication that feeding high levels of zinc for as long as 1 yr accounted for any increased incidence of tumors in mice.[1726]

SUBCELLULAR EFFECTS

Mitochondria

Zinc inhibits respiration of isolated liver mitochondria at concentrations of about 10^{-5} M.[752] Smaller concentrations induce mitochondrial swelling.[245] Effects of zinc ions on respiratory chain enzymes are complex and depend upon the particular concentration. The most sensitive reactions appear to be nicotinamide adenine dinucleotide (NAD) reduction with succinate and the electron transfer between cytochromes b and c_1. Higher concentrations of Zn^{+2} further inhibit respiration by impairing flavin and cytochrome oxidase activity.[1493]

Brierly[159] has demonstrated that Zn^{+2} stimulates energy-linked accumulation of Mg^{+2} in heart mitochondria, an action ordinarily not associated with any irreversible membrane change, implying that the uptake may be a physiologic effect.

Lysosomes

In an attempt to determine how zinc may beneficially influence various tissue injuries, Chvapil et al.[286] found that zinc stabilizes lysosomal membranes by a mechanism restricted to the surface of the membrane.

CARCINOGENESIS, TERATOGENESIS, AND MUTAGENESIS

Zinc and Cancer

Tumor induction by zinc in experimental animals. That zinc may induce tumors was first reported by Michalowsky,[1038-1040] who produced testicular

teratomas in adult roosters by injecting a 5% zinc chloride solution directly beneath the testicular capsule. Tumors developed in only a small percentage of cases. However, spontaneously occurring testicular teratomas occur only rarely.[982] Zinc was found to induce testicular tumors in fowl only if the testes were injected during the months (January-March) when the gonads were active, but they could be induced at other times of the year if the testes were stimulated by prolonged injection of gonadotropins.[64]

Injecting testes of Japanese quail with zinc chloride caused few teratomas in organs where gonadal growth was stimulated by manipulation of the photoperiod.[589] Zinc will not induce tumors in immature testes. Zinc-induced teratomas do not metastasize,[64,232] and are believed to be derived from anlagen originating in germ cells and resembling reticulum cells. These rests may become trapped in the zinc-induced scars.[1506] Zinc-induced teratomas from testes of fowl have been transplanted successfully into subcutaneous tissues and intra-abdominal cavities of other fowl.[39] The transplanted tumors grow rapidly and may invade adjacent skeletal muscle. Injection of other fibrosing substances or irritants have not produced testicular teratomas,[1038,1040] so that zinc <u>per se</u> applied in this manner somehow must provide the tumorigenic stimulus to these cells. Similar tumors may be induced in mammals. Testicular tumors have been produced in rats by intratesticular injection of zinc chloride.[1350]

Except for the ability to induce testicular teratomas, no experimental evidence exists that zinc administered orally or parenterally is tumorigenic.[1573] Mice fed diets containing 5,000 ppm zinc as zinc oleate for 3 mo, reduced to 2,500 ppm for 3 mo and then 1,250 ppm for a total time of 1 yr were studied for tumor incidence at 45 wk of age.[1726] Dosage was reduced

because of onset of anemia, weight loss, and death. Hepatoma, malignant lymphoma, and lung adenoma were found in control and experimental groups. Only the number of hepatomas exceeded the number of similar tumors in the control group (7 out of 23:3 out of 24), but these numbers were not believed to be significantly different. Addition of zinc sulfate to drinking water at 5,000 ppm and 1,000 ppm for the same period of time did not increase the incidence of tumors.[1726]

Inhibition of tumor growth by zinc. Several studies of experimental animals and observations of humans suggest that the administration of zinc may inhibit tumor growth. The initial suggestion that zinc might have such an effect arose from studies by Bishchoff and Long,[118] who injected virgin Marsh-Buffalo mice aged 2 mo with a predictably high occurrence of spontaneous mammary adenocarcinoma. Seventy-three percent of the control mice in this study developed tumors at age 15 mo, whereas tumors occurred in only 38% of the zinc-injected mice. Similarly, supplementing drinking water with zinc sulfate reduced the incidence of 9,10 dimethyl-1,2-benzanthracene-induced tumors in the cheek pouches of golden hamsters.[1268] Zinc chloride paste has been found useful in controlling local effects of inoperable breast cancer.[1524]

Tissue zinc levels in humans with malignant disease. There is no available evidence at the present time to suggest that zinc deficiency *per se* has any etiologic role in human cancer. However, a number of studies have been conducted which indicate change in tissue content of zinc in persons with malignant disease, but it is not possible now to interpret the significance of these changes. Furthermore, results from many of the studies to date appear contradictory. Reasons for the present uncertanties are manifold,

but variations in analytic methods and cytologic criteria guiding selection of material to be analyzed may be responsible for some of the difficulties.[1655]

An early report suggested that some neoplasms contain particularly high concentrations of zinc,[336] but it is clear now that this does not apply to all cancers. In fact, the converse is more generally true. Tumors usually contain lower levels of trace metals such as zinc than do non-neoplastic tissues in the same patients,[1616,1807] but exceptions may exist for specific tumors. For example, scirrhous carcinoma of the breast and bronchogenic carcinoma may contain increased amounts of zinc.[1090] But measuring zinc content of pleural fluid cannot distinguish benign and malignant lung diseases.[385]

Prostatic tissue has a particularly strong affinity for zinc,[*584,965,994,1652] and hyperplastic prostate gland significantly increases in zinc content, whereas carcinomatous portions of prostate decrease in zinc.[138,355,593,709,993,1368,1443]

In 1959, it was shown that the zinc level of blood from patients with cancer generally is subnormal;[7] and it has been suggested that the lower serum zinc levels reflect lower levels of red cell carbonic anhydrase.[8] Davies[360] found that 75% of all patients with carcinoma of the bronchus had plasma zinc levels below the normal range. In a larger study of more than 100 patients with bronchial carcinoma of all histologic types, he concluded that most patients with bronchogenic carcinoma have persistent low serum zinc and that 25% of patients with malignancies that do not metastasize have lower than normal plasma zinc. Patients with cancer at other sites do not have low plasma zinc unless they have experienced marked weight loss or have low serum protein levels.[360] Others warned that zinc metabolism is inhibited by excessive calcium, cadmium, copper and probably other elements, so that

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* See also Chapter 7.

lower plasma zinc levels can only be interpreted when amounts of other trace metals in plasma are determined simultaneously.[1556]

Lower than normal plasma zinc levels have also been observed in children with untreated leukemia. Plasma copper levels are increased but no correlation was found between copper:zinc ratios and total white cell or peripheral blast cell counts.[16] The zinc content of noncancerous portions of liver containing malignant tumor is higher than normal;[1171,1807] in one case of acute lymphatic leukemia with hepatic infiltration, the liver showed a 258% increase in zinc, a 301% increase in iron, and a 233% increase in cobalt.[1171] Elevation of hepatic zinc levels in persons dying of malignant disease not involving the liver has been found in more recent studies.[566,1807] Morgan et al.[1079] reported increased liver and kidney zinc in patients with carcinoma of the lung. Three explanations for the increase in hepatic zinc that accompanies malignancy have been postulated:

- the rise in liver zinc could reflect widespread premalignant change not peculiar to liver;
- the rise in liver zinc could be related to the poor nutritional state of patients with cancer; and
- the rise in liver zinc could be a feature of a chemical defense reaction of normal liver to invasion by malignant cells.[566]

Table 11-1 shows the results of the study by Griffith et al.[566] that confirmed the increase of liver zinc in apparently normal tissue in subjects dying from cancer. No comparable elevations occurred in kidney, heart, spleen, or pancreas; thus the authors suggested that the first two explanations were improbable.

TABLE 11-1

Tissue Zinc Levels in Subjects with and Without Malignant Disease[a]

Tissue[b]	Malignant Disease		Control Series		Probability
	Mean	Standard Deviation	Mean	Standard Deviation	
Liver	837	204	538	95	<0.05
Kidney	502	154	505	106	>0.49
Heart	301	98	364	69	>0.25
Spleen	169	64	196	42	>0.25
Pancreas	263	104	291	126	>0.30

[a] Data from Griffith et al.[566]

[b] Zinc values in mg/100 g ashed tissue.

Other explanations of the relationship between zinc metabolism and cancer involve the influence of zinc on RNA and DNA metabolism[1757] and chelating ability of chemical carcinogens.[516]

Zinc Teratogenesis and Mutagenesis

No evidence exists that excessive zinc produces any teratogenic effect. It is of interest, however, that simultaneous administration of zinc with cadmium salts will dramatically reduce the teratogenic effect of cadmium in golden hamsters.[466]

No literature was found to suggest that zinc is mutagenic.

ZINC AND CADMIUM

Introduction

The close relationship between zinc and cadmium makes it necessary to discuss both metals whenever one of them is under survey. This imperative was recognized early. In an extensive study published in 1926 on workers exposed to zinc[84] the authors pointed out that they had chosen a smelter treating zinc ore with low content of cadmium and lead to avoid significant effects from these other metals. However, this aspect has often been neglected since. Reports on effects of zinc inside or outside zinc smelters have sometimes not taken into account that significant exposure to cadmium, lead, or arsenic was also likely. Similarly, food poisoning reported to have been caused by zinc actually may have been caused by cadmium leaking from zinc-plated materials. Zinc compounds often contain relatively large amounts of cadmium, and since reports on animal exposures to large amounts of zinc seldom state the concentration of cadmium or lead, it cannot be ignored that some effects may have been caused by other metals. Aspects of relationships between zinc and cadmium have been discussed by Schroeder *et al.*[1451]

Metabolism and Effects of Cadmium

Because the metabolism of zinc has been discussed elsewhere, only the metabolism and effects of cadmium will be mentioned. For further details, two recent reviews[505a,507] should be consulted. The following section is based on those two reviews.

The systemic absorption of ingested cadmium in healthy human beings is about 6%. Data from experiments on rats indicate that calcium deficiency may cause higher absorption of cadmium. The placenta constitutes an effective

barrier to cadmium and thus the newborn is virtually free from it.

Absorbed cadmium will mainly accumulate in liver and kidneys; in normal people, the kidneys will contain about one-third of the total body burden. The main part of the cadmium is bound to a low molecular weight protein, metallothionein, which contains both cadmium and zinc. The synthesis of this protein is stimulated by exposure to cadmium. It has been claimed that zinc also may stimulate the synthesis and that metallothionein is important to zinc metabolism.[1741]

The accumulation of renal cadmium may eventually cause renal tubular dysfunction. The critical level, that is, the concentration at which the first signs may appear in sensitive individuals, has been estimated to be about 200 μg cadmium/g wet weight in renal cortex.[507] Present mean concentrations in European and North American populations at age 50 are 15-30 μg cadmium/g wet weight, whereas in parts of Japan regarded as nonpolluted by cadmium, higher values have been reported.

The first known sign of renal dysfunction in human beings is increased excretion of low molecular weight proteins. This sign may be followed by more advanced proteinuria, glycosuria, aminoaciduria, and disturbances in renal handling of calcium and phosphorus. Itai-itai disease,* found in Japan, is thought to be caused by disturbed mineral metabolism; women with calcium deficiency and renal damage caused by cadmium after prolonged ingestion of rice contaminated by the metal have exhibited the disorder.

Excretion of cadmium is very slow and thus the biologic half-life is extremely long: estimates from between 10-30 yr have been postulated.

*Itai-itai or "ouch-ouch" disease got its name from the severe pain caused by the multiple fractures of bones that occur. It is resistant to treatment with vitamin D_1, except in extremely large doses. The disorder was reported in Toyama, Japan, where rice fields had been contaminated by river water used for irrigation. The pollution source was a zinc mine many miles upstream.

INTERACTIONS BETWEEN ZINC AND CADMIUM

This section will mainly deal with *in vivo* interactions between zinc and cadmium, as a vast literature on *in vitro* effects of cadmium on zinc, especially in enzyme systems, is easily available. For accounts of such experiments, a review by Vallee and Ulmer[1663] may be consulted.

Animals

Major work in this field was initiated by Parizek,[1206] who observed that a large dose of a zinc salt could prevent the action of a large single dose of injected cadmium on the testes. His finding has been confirmed.[579, 581-583,817,818,983] Also, the teratogenic and carcinogenic effect of large injected doses of cadmium can be unequivocally prevented by zinc.[465,576] However, these were all acute experiments, involving large amounts of injected cadmium and zinc and with little application to long-term toxicity problems.

That dietary cadmium could accentuate zinc deficiency symptoms was first shown in turkeys.[1575] The adverse effect of cadmium could be reduced by increasing zinc intake. Similar findings were then obtained in chicks.[1574] This research had great heuristic value. Groups of chicks were fed diets with varying amounts of zinc and cadmium (and effects of copper and iron were studied as well).[703] The basic diet contained 25 μg zinc/g and was supplemented with 200 μg zinc/g and/or 100 μg cadmium/g. It was found that cadmium depressed the growth rate and caused pathologic gizzard changes and that both these changes were reversed by zinc. Cadmium brought about hypochromic anemia, which was not reversed by zinc. Bunn and Matrone[201] studied groups of rats and mice. Some groups were pretreated so that they were copper-deficient. Zinc was added to the diet, which contained 9 μg zinc/g and 2 μg copper/g in a concentration of 200 or 400 μg zinc/g diet and/or cadmium in a concentration of 100 μg/g diet for 5 wk.

Whereas supplemented zinc did not increase zinc concentrations in liver or testes, ingested cadmium caused zinc concentrations to increase significantly in these organs in normal and copper-depleted mice on both basic and supplemental zinc diets. Also, supplemental copper increased zinc concentrations, especially in the testes. In rats the results were not so consistent. Cadmium exposure actually decreased liver zinc in copper-depleted animals on the basal diet, but hepatic zinc markedly increased in animals pretreated with a commercial diet.

Even in zinc-supplemented and copper-depleted animals, there was only a slight increase in liver zinc, whereas animals treated with copper plus zinc increased their zinc levels markedly after cadmium exposure. Cadmium caused anemia, which was partly reversed by copper, but not by zinc. Indeed, anemia was more pronounced in mice given both zinc and cadmium.

Banis et al.[70] made similar studies on rats. Zinc (200 µg/g), cadmium (100 µg/g), and iron (68 µg/g), alone or in combination, were added to a basic diet. Cadmium depressed weight gain, and although zinc or iron alone did not reverse this effect, zinc plus iron did. Cadmium decreased hemoglobin concentration and cadmium plus zinc further reduced it. Another experiment demonstrated that cadmium intake increased liver zinc in animals on basal diets.[70]

Miller et al.[1059] found increased fecal zinc in calves given a diet with 40 µg zinc/g and 350 µg cadmium/g. Powell et al.[1269] found that in calves given 640 µg cadmium/g in a diet that also contained 27 µg/g of zinc, kidney and liver levels of zinc increased. No tissues examined were found with zinc levels below those of the controls.

That there was an increase in fecal zinc after ingestion of cadmium seems to conflict with the findings of Gunn et al.,[578] who found that injecting cadmium in rats caused reduced excretion of zinc-65 via feces. However, the high amount of dietary cadmium given by Miller et al.[1059] may well have reduced zinc absorption. The results of Gunn et al.[578] will be discussed in detail later.

Pond et al.[1259] gave 154 µg cadmium/g diet to pigs on diets high (74 µg/g) and low (22 µg/g) in zinc for 6 wk. Whereas low zinc alone did not depress weight gain, ingestion of cadmium did cause a significant drop, although the effect was partially overcome by the higher zinc intake. Fox et al.[504a] found that in young Japanese quail given a diet with cadmium (75 µg/g) and zinc (75 µg/g), cadmium markedly reduced the zinc content of the tibia.

All the above experiments were performed using high dietary levels of cadmium and/or zinc. In contrast, Petering et al.[1236] gave groups of weanling rats a zinc-deficient diet (<2 µg zinc/g) and zinc in drinking water in concentrations of 0, 2, 8, and 32 µg/g. Optimum growth was obtained with 8 µg/g, whereas at 2 µg/g, growth was marginal. After 4 wk, 50% of the groups on 2 and 8 µg zinc/g water were then given cadmium in the water at a concentration of 3.4 µg/g for about 7 wk. The molar ratios of zinc:cadmium were 1:1 and 4:1. The cadmium exposure was considerably lower than those used earlier, and it is more realistic. It was found that in the group on 2 µg/g water, cadmium decreased the growth rate, and was responsible for some pathologic changes, such as corneal keratinization, which are seen in zinc-deficient animals. Gonads did not seem to be affected. Rats on higher zinc intake were not influenced by cadmium.

The most significant finding with regard to distribution of zinc was that the zinc concentrations in testes markedly decreased in rats given marginal zinc and cadmium (molar ratio, 1:1), whereas no such change was noted

in rats given zinc:cadmium at a ratio of 4:1. Therefore, cadmium can cause changes in distribution of zinc at marginal intakes. Zinc will still be stored in organs such as liver and kidney, where cadmium will accumulate. However, zinc may be depleted in other organs where cadmium does not accumulate as greatly.

Most studies on long-term interactions between zinc and cadmium are ingestion experiments, and few studies use injection techniques. Vigliani[1691] reported on rabbits given subcutaneous injections of 0.25 mg cadmium/kg body weight, 5 days a week for several months. Some animals also were given injections of equimolar amounts of zinc. Morphologic changes and degree of proteinuria revealed that cadmium alone caused more severe renal tubular damage than cadmium plus zinc.[1691]

That cadmium can alter the metabolism of zinc has also been shown in isotope experiments using zinc-65. Cotzias et al.[322] gave 63 uM of cadmium, zinc, copper, or mercury as the sulfate or citrate in a single intravenous injection to rabbits, alone, simultaneously with, or after an injection of zinc-65 as the chloride. Cadmium as well as stable zinc changed the clearance curves of zinc-65 in blood similarly, whereas the other metals had no effect. And, there were differences in the distribution of zinc-65 activity in organs after the administration of zinc or cadmium. The authors interpreted the results to mean that some degree of interchange between cadmium and zinc occurred in vivo, but zinc-zinc and cadmium-zinc interchanges were not identical.

When zinc or cadmium was given some time after the zinc-65 injection, differences appeared in the plasma zinc-65 curves, indicating a difference in exchange. The doses administered were high, but nevertheless these data indicate a possible interaction between cadmium and zinc.

Gunn et al.[577] found that cadmium decreased the uptake of zinc-65 in the testes and dorsolateral prostate. They conducted the following four studies on rats and mice:[578]

- Rats were given an intracardial dose of zinc-65 4 mo, 1 wk, and 1 day after a subcutaneous injection of 3.4 mg cadmium/kg body weight. After 24 h, the zinc uptake of organs was measured. Increased zinc uptake was noted in liver, pancreas, and kidney, whereas no difference was found in skeletal muscle.
- After an intracardial injection of zinc-65 to rats, an intracardial injection of 2 mg cadmium/kg body weight was given 24 h later. Liver content of zinc-65 was measured 5 and 48 h after the cadmium injection. Five hours after injection, concentration of zinc-65 decreased in cadmium-treated animals compared to saline-treated controls whereas after 48 h the reverse was found. This initial displacement of zinc from the liver had been noted before.[322]
- Zinc-65 was given to rats in an intracardial injection and feces were collected daily. Four days after the zinc dose, half of the rats were given a subcutaneous injection of 3.4 mg cadmium/kg body weight. Feces were collected for another 3 days. Compared to controls, zinc excretion decreased in the animals given cadmium and it was found that zinc-65 concentrations were higher in the cadmium-treated animals.

- Three days after mice were injected subcutaneously with 3.4 mg cadmium/kg body weight, they were given a subcutaneous dose of zinc-65. Animals were killed 1, 2, 3, and 7 days after the zinc-65 injection. Activity was measured in liver, kidney, pancreas, and carcass. Whole body retention was 15-20% greater in cadmium-treated rats than in controls.

The authors interpreted the results to mean that cadmium was interfering with the gastrointestinal excretion of zinc, thus causing zinc levels to rise in liver, kidney and pancreas. The present knowledge of cadmium metabolism suggests the opposite, that is, the decrease in excretion was caused by increased zinc retention.

Studies also have been conducted on more specific physiologic problems. Whereas large doses of injected cadmium bound to cysteine increased renal tubular reabsorption of sodium, zinc had no effect at all.[1677] Cadmium, either after injections or long-term, low-level ingestion, can cause hypertension in experimental animals; it occurs at a molar cadmium:zinc ratio of > 0.4.[1445] When a zinc chelate was injected into animals made hypertensive by cadmium, blood pressure decreased rapidly.[1445]

There are a number of zinc-dependent enzymes which could be susceptible in vivo to cadmium interference. Alkaline phosphatase activity decreased in renal cortex of rabbits exposed to cadmium for 6 mo by subcutaneous injections.[60] This decrease still existed 6 mo after the last injection. It was postulated that leucine aminopeptidase would be a kidney enzyme susceptible to cadmium.[1691] This enzyme is important for protein catabolism in the kidney and interference with that system could be influential in the development of tubular proteinuria. Cousins et al.[324] showed that in cadmium-exposed pigs the activity of this enzyme in renal cortex was decreased after 150 µg cadmium/g in the diet (80 µg zinc/g) for 6 wk, resulting in a mean renal concentration of 78 µg cadmium/g wet weight, and about 100 µg/g wet weight in renal cortex.

Because different levels of cadmium were given and both cadmium and zinc were determined in organs, the research also dealt with distribution of zinc

in relation to cadmium. Zinc levels increased in whole kidney, and on a molar basis there was a distinct change between 150 and 450 µg cadmium/g diet. Exposure to 50 and 150 µg cadmium/g resulted in molar cadmium:zinc ratios of 0.55 and 0.71 respectively, whereas 450 µg/g made a molar cadmium:zinc ratio of 2.73. The renal concentrations of cadmium were 41, 78, and 276 µg/g wet weight. These results indicate that the critical level in this experiment must have been between 100 and 300 µg/g wet weight in renal cortex. The decreased leucine aminopeptidase activity indicates that the level could well be close to 100 µg/g wet weight.

Liver enzymes such as glutamic-oxaloacetic transaminase (GOT), alkaline phosphatase, aldolase, and succinoxidase, as well as oxidative phosphorylation, were studied by Sporn et al.[1539] Young rats were given diets supplemented with 10 µg cadmium/g and/or 80 µg zinc/g. Growth rate was not affected by cadmium. Cadmium caused a decrease in phosphorylation, but zinc counteracted this effect. Cadmium also reduced the activity of GOT, alkaline phosphatase, aldolase, and the succinoxidase system, but zinc did not affect this change. The authors emphasized that the zinc-cadmium interaction was not a case of general antagonism, but an effect limited to certain enzymes and metabolic sequences. Original cadmium and zinc levels were not reported. It was calculated by using data from similar exposures that the amounts of liver cadmium could only have been a few µg/g wet weight.

Long-term effects of cadmium were studied in two generations of mice.[1449] Cadmium was given in drinking water at a concentration of 10 µg/g from the end of the weaning period until the end of the experiment. The metal was found to be toxic for breeding mice to such an extent that there was no

survival beyond the second generation. Congenital abnormalities appeared at a much higher frequency than in controls. Cadmium does not traverse the placenta and it is conceivable that a teratogenic effect was caused by a secondary zinc deficiency in the fetuses. However, cadmium and zinc were not determined in the organs.

A recent paper on normal levels of cadmium and zinc in horse kidneys[1249a] reported that cadmium concentrations in renal cortex varied from 5-250 µg/g wet weight in 37 horses aged 1-25 yr. At lower cadmium concentrations (5-70 µg/g) an equimolar increase in zinc occurred, but at higher cadmium concentrations zinc did not increase to the same extent as cadmium.

Humans

Information on the relationship between cadmium and zinc in humans has been obtained by autopsy studies. Schroeder et al.[1451] showed that parallel increases in zinc and cadmium levels in kidney occurred with age. Piscator and Lind[1250] found the same relationship and demonstrated that the increase in zinc was equimolar to the increase in cadmium. This discovery was thought to be connected to the presence of metallothionein in the renal cortex, because this protein normally has equimolar amounts of the two metals. Furthermore, it was shown that if the amount of zinc equimolar to the cadmium were subtracted from the total zinc, the physiologic amount of zinc in the cortex could be determined. The concentration of this zinc fraction was found to be about 160 µg/g dry weight (34 µg/g wet weight). Piscator and Lind's data were obtained from members of the general population. They were not victims of occupational exposure, and had concentrations of cadmium below levels that could cause renal damage.

CHAPTER 12

STANDARDS FOR ZINC LEVELS

EXISTING STANDARDS FOR AIR QUALITY

Air quality standards for zinc and its compounds have been established in many countries for occupational exposures, but a thorough search of the literature did not reveal any standards for public exposure.

Table 12-1 presents the current standards for industrial situations, all of which are maximum allowable concentrations (MAC).[771] Their variations may arise partly from historical reasons. Before 1962, the U.S. standard for zinc oxide fume was 15 mg/m^3 and was based primarily on the work of Drinker[402a] and Fairhall.[451a] Subsequent experience showed that zinc fume fever occurred from exposures to less than 15 mg/m^3.[28] No adverse effects have been reported where this standard is observed.

EXISTING STANDARDS FOR WATER QUALITY[28b,1011b,1645b,1806a]

Standards for drinking water quality for zinc are based primarily on esthetic characteristics such as taste and cloudiness. Most standards recommend 5 mg soluble zinc/l water, which is the threshold for the astringent taste. At 30 mg/l the water becomes cloudy, and at 40 mg/l it has a metallic taste.[829] Acute gastrointestinal distress will occur from concentrations in the range of 280 mg/l and above.

Zinc in Drinking Water

The levels at which zinc is found most frequently in drinking waters cannot be considered to be detrimental to human health. The current recommended standard is 5 mg/l. Zinc is an essential trace element in human and animal nutrition with a recommended daily allowance of 15 mg/day for adults and 10 mg/day for growing children.[1119]

TABLE 12-1

Occupational Standards for Zinc and Zinc Compounds in Air[a]

Country	Compound	Level	Comments
Bulgaria	Zinc oxide	10 mg/m^3	[b]
Czechoslovakia	Zinc oxide	5 mg/m^3	MAC
Finland	Zinc oxide fume	15 mg/m^3	MAC
Germany (entire)	Zinc oxide fume	5 mg/m^3	MAC
Hungary	Zinc oxide	5 mg/m^3	MAC
Japan[789b]	Zinc oxide fume	5 mg/m^3	MAC
Poland	Zinc oxide	5 mg/m^3	MAC
Romania	Zinc oxide fume	10 mg/m^3	MAC
Romania	Zinc pentachlorothiophenate	5 mg/m^3	MAC
Sweden[1790a]	Zinc oxide	5 mg/m^3	MAC
United Arab Republic Syrian Arab Republic	Zinc oxide fume	15 mg/m^3	MAC
USA, entire	Zinc chloride fume	1 mg/m^3	MAC
USA, entire	Zinc oxide fume	5 mg/m^3	MAC
USA, Pennsylvania	Zinc oxide fume	10 mg/m^3	MAC[c]
USA, Massachusetts	Zinc chromate	0.2 mg/m^3	MAC
USSR, entire (1975)[1363a]	Zinc oxide	6 mg/m^3	MAC
USSR, entire (1972)	Zinc pentachlorothiophenate	2 mg/m^3	MAC
World Health Organization	Zinc oxide	5 mg/m^3	MAC
Yugoslavia	Zinc oxide	5 mg/m^3	MAC

TABLE 12-1 - continued

[a]Unless otherwise noted, data from ILO/WHO Committee on Occupational Health[771] and the American Conference of Governmental Industrial Hygienists.

[b]Maximum allowable concentrations, the equivalent of threshold limit value, are the time-weighted average concentrations to which nearly all workers may be exposed for eight hours a day, five days a week, for their working lifetime without expectation of any adverse health effects.

[c]At this level, the maximum exposure could be no more than 30 min.

In a survey by EPA, a nationwide water analysis of 591 samples failed to find any sample that exceeded 4 mg/l.[1646b] Craun and McCabe reported that the concentration of zinc at treatment plants was well below the current standard of 5 mg/l.[332a] However, in cities with soft, acidic water, such as Seattle and Boston, pickup of zinc occurred from the distribution system between the treatment plant and the tap. In Boston, 35% of 108 samples showed such pickup, but both mean and maximum concentrations in running tap water were within the current limit (223 and 1,625 µg/l, respectively). In the more acidic waters of Seattle, zinc pickup was found in 95% of samples and 10% exceeded the standard of 5 mg/l. The maximum concentration found was 5.46 mg/l.

Although zinc can be acutely toxic when inhaled, there is little evidence of acute oral toxicosis. Two adults reportedly drank water containing 40 mg/l and experienced irritability, muscular stiffness and pain, loss of appetite, and nausea.[1119a] There are, however, other reports of individuals drinking water with zinc concentrations from 20-50 mg/l with no adverse effects.[35,83,707a]

Zinc salts act as gastrointestinal irritants. Although the illness is acute, it is transitory. The concentration range in water at which zinc will be an emetic is 675-2,280 mg/l. In tests performed by a taste panel, 5% of the observers were able to distinguish between water containing 4 mg/l (when present as zinc sulfate) and water without zinc salts. The water was described as having a bitter or astringent taste.[297]

Zinc is known to interact with other trace metals. It has been shown to have a protective action against cadmium and lead toxicoses. Animal data suggest that the zinc:copper ratio in the diet may be important and as noted above there also appears to be a relationship between dietary zinc and iron.[1144b,1403b] Therefore, it may be possible that the ratios of these trace metals in drinking water may have as much significance as the individual levels.

Cadmium and lead are common contaminants of zinc used in galvanizing. Assuming that zinc is dissolved from galvanized water pipes no less than cadmium, dissolution of zinc to produce 5 mg/l would be accompanied by something less than the allowable 0.01 mg cadmium/l when cadmium contamination of the zinc is as high as 0.03%. The cadmium may contribute to the acute gastrointestinal distress following ingestion. Similarly, lead concentrations would likely be increased by something less than the allowable 0.05 mg/l when lead contamination of the zinc reaches as high as 0.6%. Based on the available evidence, the current drinking water standard of 5 mg/l provides sufficient protection for consumers.

Zinc in Waterways

Kopp and Kroner[869] reported that in 1,207 positive tests for zinc on samples from U.S. waterways, the highest observed value was 1,138 µg/l (from the Cuyahoga River at Cleveland, Ohio) and the mean was 64 µg/l. Soluble zinc was measured in over 76% of all water samples tested. The highest mean zinc value, 205 µg/l, was found in the Lake Erie basin, whereas the lowest mean zinc value, 16 µg/l, was observed in the California basin. In seawater, the highest concentration of zinc has been found to be about 10 µg/l.

The toxicity of zinc compounds to aquatic animals is modified by several environmental factors, particularly hardness, dissolved oxygen, and temperature. Skidmore,[1490] in a review of the literature on the toxicity of zinc to fish, reported that salts of the alkaline earth metals are antagonistic to the action of zinc salts, and salts of certain heavy metals are synergistic in soft water. Elevations in temperature and reductions in dissolved oxygen increase the toxicity of zinc. Toxic concentrations of zinc compounds can affect the morphology and physiology of fish adversely. Acutely toxic concentrations induce cellular breakdown of the gills, and possibly clog the

gills with mucus. Conversely, chronically toxic concentrations of zinc compounds bring about general enfeeblement and widespread histologic changes to many organs, but not gills. Growth and maturation are retarded.*

*See Chapter 6, "Zinc in Aquatic Animals," for a detailed discussion of the effects of zinc on fish.

CHAPTER 13

SAMPLING AND MEASUREMENT TECHNIQUES FOR ANALYZING ZINC

SOLID AND LIQUID SAMPLES

Although the analytic chemical literature pertaining to zinc is quite extensive, only a limited number of techniques have enjoyed the benefit of extensive evaluation by various organizations of analysts and, where possible, these procedures are emphasized. Attention is paid to the evaluation of zinc in physiologic fluids and other biologic media, potable and polluted waters, foodstuffs, and soils and plants.

Techniques for Handling Samples

Levels of zinc in most media of direct interest to environmental health occur at the trace level and regardless of measurement technique, require that maximum care be exercised to minimize both loss of zinc from the sample and the contamination of the metal by zinc-containing reagents, vessels, etc. Contamination is especially worrisome in the case of zinc because of its ubiquitous presence in the general environment. Details of sampling procedure to minimize the above problems have been fairly well standardized and one is directed to the discussion by Thiers[1606] for specific information.

In most procedures for zinc analysis, some prior treatment of the sample is necessary and involves destruction of the zinc-containing sample matrix. In practice, this involves wet-, dry-, and low-temperature ashing. A detailed discussion of the merits and disadvantages of the various ashing techniques may be found in a recent book by Gorsuch.[551]

Zinc in Biologic Media

Unlike procedures for foodstuffs, soils, plants and waters, standardized official methods for zinc analysis in physiologic media have not evolved.

Consequently, the limits of accuracy, precision and detection which have been developed in representative methods will be noted.

With liquid samples, deproteinization is usually carried out by using denaturants such as trichloroacetic acid, hydrochloric acid, [1283,1369,1753] or a combination of both. Mineralizing techniques involving wet or dry ashing may also be carried out and these pretreatments are invariably necessary for most zinc analyses of bone, [1140] [642] hair, [661,809,1656] and soft tissue. A newer, highly efficient mode of dry ashing is low-temperature ashing, [541] in which organic material is removed from the sample by an oxygenated high-energy plasma generated in a combustion unit by a radio-frequency discharge.

Most spectrophotometric procedures for zinc determination in biologic media have involved the use of chelating agents which form colored coordination complexes with divalent zinc. [1044] The most common is diphenylthiocarbazone (dithizone) and zincon. Dithizone is only moderately specific for zinc complexation and requires the concomitant use of masking agents and/or steps for removing interferents, whereas zincon is more selective to zinc complexation but forms a highly labile and complex chromophore which necessitates rapid analysis.

Measurement of zinc in biologic media by atomic absorption spectrometry (AAS) appears to be the current method of choice in most laboratories. Sample handling and reagent use steps are greatly reduced while sensitivity, accuracy, and precision remain superior. With the advent of microsampling techniques in AAS, such as the Delves cup [373] and

boat [811] techniques,* as well as flameless atomic absorption spectral techniques [650] --sampling manipulation and lower detection limits for elements like zinc are being revised downward continually, as is apparent from the literature. Fluid samples may be deproteinated and diluted [1162,1170] or simply diluted: [1031] both approaches enjoy routine use. Because they may enhance or repress the zinc absorption signal, chemical and physical matrix effects should be considered in zinc assay by AAS, although appropriate steps can be taken to minimize them. [278] When bone, hair, or soft tissue is involved, wet or dry ashing is necessary for conventional atomic absorption spectral analysis. Further, it is advisable to dry ash samples in platinum crucibles, as porcelain crucibles have been shown to entrap zinc on their surface. [624] For hair samples, an analytic compromise usually must be reached between chemical cleaning of hair surface to remove contaminating metals and leaching of zinc from hair by vigorous cleaning.

A recent text of clinical chemistry techniques describes recommended methods for zinc evaluation in serum and urine. [1750] Serum is deproteinized with trichloroacetic acid, and the resulting supernatant is analyzed directly by AAS using a zinc 214 nm line. To deproteinize urine, zinc is chelated with ammonium pyrrolidino-dithiocarbamate and extracted with methyl isobutyl ketone,.

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*The Delves cup method is a microanalytic procedure for volatile elements such as lead, cadmium, etc. The sample is placed in a nickel crucible, which is inserted in the flame portion of an atomic absorption spectrometer. The burner is equipped with an absorption tube to enhance the collection of the atomized element present. The tube is fabricated of a ceramic or quartz material. With the boat technique, samples are placed in shallow boats constructed of tantalum and inserted into the flame portion of the instrument.

followed by aspiration of the organic phase into the flame of an atomic absorption spectrometer.

Neutron activation analysis (NAA) appears to be the method of choice where very high sensitivity is desired and the requisite highly sophisticated instrumentation is not available. Unlike other popular techniques, activation analysis permits determination of many trace metals or metalloids. Examples of the application of this method to zinc determination in biologic media are available.[469,1165,1210]

Emission spectrography, using direct (dc) or alternating current (ac) arcs for excitation, is a multielement technique that has been employed for several years to measure zinc and other elements. But this technique has been limited by its lack of precision in exacting quantitative work, although this drawback is being offset by the use of increasingly more stable arcing accessories.[1651]

Spectrofluorometric techniques for zinc analysis, a procedure describing fluorometric analysis of zinc via the 8-quinolinol complex, demonstrates a marked fluorescence at 517 nm when excited at 375 nm.[970]

Anodic stripping voltammetry (ASV)[988] is a newer method for zinc measurement; it demonstrates excellent sensitivity but requires that considerable pains be taken in sample pretreatment. More comparative data and research on the range of media studies are necessary to help establish ASV as a competitive alternative to atomic absorption or colorimetric techniques.

In media such as soft tissue, in which various means of sampling are available, the method of expressing data is a particularly vexing

problem in zinc and other element analysis. How data are expressed has hampered correlation of data on zinc levels from various sources. Measurement of zinc on a wet weight of tissue basis is made risky because evaporative loss may be incurred. And losses will vary with time the sample is exposed to the atmosphere, removal of excess moisture with filter paper, etc. An assessment of zinc on a lyophilic dry weight basis must consider moisture uptake by hygroscopic residues. Dry ash analyses may suffer from incomplete ashing, which will influence the ash weight. Some uniformity may be introduced by expressing zinc content per unit weight of protein, DNA, or other biochemical marker.

Measuring Zinc in Potable and Waste Water

Of the various acceptable techniques for zinc evaluation in potable and polluted waters, those involving spectrophotometry were standardized and extensively employed first. They operate through colored complexes formed by complexing of zinc with selected chelating agents. Representative are the dithizone and zincon methods (see Appendix B for details) for examination of waters.[1311a] Colorimetric techniques for zinc in water are tedious procedures with problems of interference with the chromophore and stability, difficulties similar to those encountered in sampling biologic media of the generated complex.

The presently accepted procedures are polarography[1311a] and AAS,[319,1010,1023,1191b] the latter perhaps enjoying widest use. Polarographic methods for evaluating zinc itself or zinc along with other metals have been described.[1311a] Sample treatment includes preliminary removal of organic matter interferents from 100-ml samples by successive evaporations to dryness with concentrated nitric acid,

followed by final manipulation with concentrated hydrochloric acid. Samples relatively free of organic matter may be handled with a 1:15 ratio of hydrochloric acid:water. Zinc is easily measured polarographically and the method permits routine evaluation of zinc in the 0.01-0.1 mg/l range.

AAS is especially suited to the evaluation of zinc in water samples:[319,1010,1023,1191b] in many cases, direct aspiration of the samples into the burner of the spectrophotometer can be carried out. The literature about zinc measurement in waters by AAS is extensive and growing. One is referred to the annual surveys in the Journal of the Water Pollution Control Federation, biennial review of water analysis, Analytical Chemistry, and an annual bibliography of AAS appearing in the Atomic Absorption Newsletter.

Measuring Zinc in Soils

Levels of zinc in most soils are usually in the trace quantity range and are examined by the analytic techniques previously mentioned. In a standardized spectrophotometric procedure for zinc in soil[628,1382] in which the dithizonate complex is used, soil samples are ground, sieved, and digested with various mixtures of concentrated acids. Hydrofluoric acid is included when removing silicate is desired. Subsequent manipulation includes the use of citrate to mask iron and copper. Copper is removed as the dithizonate or is masked by carbamate or thiosulfate. The zinc dithizonate is extracted into an organic medium at a pH of 8.3. Polarographic measurement of the zinc content of soil digests or extracts, prepared as for the spectrophotometric procedure above, also have been described, although satisfactory results are offset by the tedium of the overall assay.[1029]

Soil zinc levels may also be obtained by AAS.[37] Soil samples are either wet-ashed or dry-ashed with a mixture of perchloric, nitric and hydrofluoric acids or extracted with either hydrochloric acid or a chelating agent into an organic medium. Using ammonium pyrrolidino-dithiocarbamate and methyl isobutyl ketone as agents of chelation and extraction permits a multifold increase in overall sensitivity because of the high aqueous:organic phase ratios as well as the enhanced element signals found in organic media compared to an aqueous milieu. An alternative to concentrating is the use of ion exchange resins, for which a seventyfold increase was reported once.

When zinc levels are desired as part of a multielement profile for a given sample, emission spectrography with an ac arc has been employed.[628] Samples are treated with a mixture of nitric, sulfuric, and perchloric acids to destroy organic matter. Silicate is removed by charring with hydrofluoric acid and a prepared buffered mixture. Aliquots are then placed in the carbon electrode crater for analysis.

Zinc has been analyzed in standard rock samples.[477,478] Zinc in rocks G-1 and W-1[478] was measured by optical emission spectrometry (OES), X-ray fluorescence (XRF) analysis, NAA, ASV, polarography, AAS, and spark-source mass spectrometry (SSMS). The values corresponded enough from the various methods to permit zinc levels to be established for G-1 (45 ppm zinc) and W-1 (82 ppm zinc). To compile data for the new rock standards G-2, GSP-1, AGV-1, PCC-1, DTS-1 and BCR-1, methods of zinc analysis included OES, AAS, XRF, NAA, and polarography, and the following means were yielded:

477,478

Rock	Mean, ppm zinc
G-2	74.9
GSP-1	143.0
AGV-1	112.0
PCC-1	53.0
DTS-1	61.0
BCR-1	132.0

Evaluating Zinc in Foodstuffs

Two recommended methods for evaluating zinc in foodstuffs involve
725 34,1357
spectrophotometry and AAS.

In the colorimetric technique, samples are wet-ashed in the usual manner with nitric, sulfuric, and perchloric acids. Elements other than zinc are eliminated in large measure as the sulfides, whereas nickel and cobalt are removed with various chelating agents. Zinc is isolated as the dithizonate complex through carbon tetrachloride extraction and spectrophotometrically measured as such.

Ten collaborating groups have evolved an atomic absorption method
1357
for zinc in foods, and it was deemed accurate and precise. Sources of zinc sampled in the study included sucrose, soybean meal, and white and whole wheat flour. Both wet- and dry-ashing steps were employed in the study. Recoveries were 98-102% with precision of 0.2-2.0% standard deviation.

NAA has also been applied to evaluation of zinc in foodstuffs. Of particular interest is the NAA program of the Food and
1597
Drug Administration, in which relatively thorough assessments are being made of the elemental (including zinc) makeup of food stuffs of major economic importance. Samples are digested with

sulfuric acid and hydrogen peroxide and in the case of foods, radiochemical separations are carried out to simplify radionuclide counting.

Analyzing Zinc in Plants

The classic standard procedure for measuring zinc in plants is the spectrophotometric method, involving a mixed or single color technique measuring zinc as the dithizonate.[725] Samples are dry-ashed at 500-550 C and the residues taken up in dilute hydrochloric acid. After a series of extraction steps to rid the analytic medium of interfering ions, zinc is then measured as the dithizonate, either in the presence of (mixed color) or in the absence of (single color) an excess of complexing agent.

AAS is rapidly becoming the most popular method of analyzing zinc in plants. Direct analysis of plant digests may be performed with little interference from other elements present. This method has been recommended for plant analysis on the basis of a comparative study between the Association of Official Analytical Chemists (AOAC) colorimetric procedure and AAS.[1507] Samples--which included corn, sorghum, wheat, and alfalfa--were wet- and dry-ashed. The accuracy of the AAS method was found to be as good as or superior to the standard colorimetric assay.

AIR

Zinc is encountered as an atmospheric ingredient entirely in suspended dust and dirt, so that determining zinc in the atmosphere becomes a matter of collecting and analyzing atmospheric particulates. Because zinc is often present in trace concentrations on the order of micrograms per cubic meter or less, the sampling and subsequent analyses require careful planning. Care is especially necessary in analytic procedures which determine zinc and 20-30 other elements simultaneously. This chapter summarizes methods of sampling airborne particulates and describes some of the advanced procedures for determining their zinc content.

Surveys and Reviews

In recent years, airborne particles have received considerable attention from investigators of air pollution, and this interest has coincided with the development of new instrumental methods of chemical analysis for zinc and other elements present in trace concentrations. Therefore, a wealth of literature exists on the collection and analysis of airborne particulates so that a comprehensive bibliographic review could easily become a treatise in itself. For the scope of this report, only the most recent and pertinent references have been cited.*

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* Chemical Abstracts (1967-1974), Air Pollution Abstracts (1971-1974), The Analyst (1970-1974), Analytica Chimica Acta (1970-1974), Analytical Chemistry (1972-1974), Talanta (1973-1974), Staub Reinhaltung der Luft English edition (1972-1974), Atmospheric Environment (1971-1974), Applied Spectroscopy Reviews (1969-1974), and the Journal of Radioanalytical Chemistry (1971-1974) were examined.

The sampling and chemical analysis of atmospheric particulates have been the subject of surveys, reviews, and collections of papers [439, 772, 1030, 1489] which do not always describe zinc determinations specifically, but are useful nonetheless for providing an overview of particle collection and characterization against which the occurrence and determination of zinc can be related.

A procedures manual by the Atomic Energy Commission (AEC)[1644] reviewed air sampling and elemental determinations, including zinc determined by AAS. West[1758] reviewed the trace metal analysis of inorganic particulates and concluded that classical methods of gravimetric and titrimetric analysis are being replaced by instrumental methods offering advantages of selectivity and sensitivity: colorimetric and spectrophotometric methods, ring oven methods, emission spectrography, flame photometry, AAS, and polarography. Greifer and Taylor[564] surveyed methods for determining trace elements (concentrations less than 100 ppm) in environmentally important materials such as coal, fly ash, and incinerator ash that could eventually find their way into the atmosphere as particulates. They described the determination of zinc and 26 other elements using nuclear methods, SSMS, XRF and X-ray emission, absorption spectrophotometry, atomic emission spectrography, voltammetry (polarography), and potentiometry (ion-selective electrodes). Kane and Larrabee [815] reviewed trace analysis techniques for solids and their treatment of emission spectrography, activation analysis, SSMS, and XRF is directly applicable to particulate analysis for zinc content. Zief and Speights [1818] reviewed the techniques of trace element analysis and described sample handling and analysis by emission spectrography, flame spectrophotometry, absorption spectrophotometry, neutron activation, and coulometry. Specific information on zinc determinations was included where applicable.

Atmospheric Sampling

To determine zinc in the atmosphere, airborne particulates must be collected and transported to the laboratory for chemical analysis because no good methods exist for carrying out trace element analysis in the field. Atmospheric sampling involves careful planning to assure that the elemental content of the samples in the laboratory is truly representative of the atmosphere in the locations sampled. Careless sample collection and handling may invalidate the most sophisticated and accurate chemical analyses.

The number of samples taken for analysis and the variety of locations sampled must be sufficiently great to assure that all the trace elements, and zinc in particular, be represented in their correct concentrations in the collected specimens. Caution must be exercised to avoid problems arising from inhomogeneity of the particulates, variation of composition during sampling, sample alteration during transport, i.e., zinc losses or zinc contamination, and inefficient collectors which do not capture all the trace elements. A full treatment of the best places to sample, the best times, how big a sample to take, how often and for how long, and how to collect and retain the zinc and other trace elements is outside the scope of this chapter. These matters are essential to a proper interpretation of the analytic data, and they may assume even greater importance if health considerations or legal implications are involved. Many of these principles of trace element sampling are discussed by Hendrickson,[664] Fair et al.,[451] and the AEC.[1644]

There are various techniques for collecting suspended particulates from the air, and the most widely used are filtration methods and impingers. Other particle collectors include electrostatic precipitators, sedimentation bottles, thermal precipitators, and liquid sorption traps.[665, 910]

The sampling techniques may be divided into two broad categories: those which collect total particulates and those which fractionate the samples into size classifications. The latter samplers are popular where the occurrence of zinc in the atmosphere is to be correlated with particle sizes.

Most particulate samples are obtained with filters, the most popular filter materials being paper (randomly matted cellulose fiber), membrane (thin plastic structures with flat, porous surfaces), and glass fiber. Metallic silver is used at times, as when wet chemical analyses require acid dissolution of the filter material. Membrane filters (Millipore, Gelman, Nuclepore, etc.*) collect on their surface (as opposed to the bulk material matrix)all particles of a size larger than the stated pore size. They are available in a variety of materials such as cellulose acetate-nitrate copolymer, polyvinyl chloride, nylon, and Teflon. The 0.45-μm and 0.8-μm pore sizes are widely used for air sampling.

Contrary to popular belief, filters do not work by simply straining the air stream, although this sieving action is one mechanism by which particles are trapped. The primary filter action is impaction, resulting when the air stream suffers a sudden change in velocity (magnitude and direction) and the particles continue onward from inertia to impact with the substrate where they remain entrapped. The relative effects of impaction versus sieving are shown by the ability of Whatman 41 filter paper (widely used in air sampling) to collect about 98% of 0.2 μm particles from an air stream of 100 cm/sec face velocity although the pore size is

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* Trade names have been identified solely to help readers and do not imply any endorsement or recommendation by the National Academy of Sciences or the National Research Council.

20 μm,[910] or the 0.45 μm pore size membrane filter to collect 0.1-μm particles.[665] Diffusion, a third filter mechanism, is of minor importance with the thin filters and the relatively high air velocities used.[910] High volume samplers, such as those used in the U.S. National Air Sampling Network (NASN) draw air through a filter at a rate of 1.1-1.7 m^3/min (1,100-1,700 l/min) and are considered a standard method of outdoor particle collection.[782] Low volume samplers such as are used for sampling the occupational environment operate in the range of 5-50 l/min; and personal (lapel) samplers, limited in size and weight to small battery-operated devices, operate at 1-3 l/min.[1644]

Impaction devices are samplers which collect particles impinging on surfaces (as cascade impactors) or in liquid bubblers (as midget impingers). Cascade impactors such as the Andersen sampler are made up of a series of stages (often six or more). The stages consist of plates containing holes or slots of progressively decreasing size, which effect a size separation of air-entrained particles. The particles are not sieved by the holes but are deposited according to the air velocities in each stage; the largest particles are deposited in the earliest stages where the air velocities are lowest, and the finer particles are carried toward the later stages where the air velocities increase with decreasing hole size. The overall volume rate of flow remains constant, about 28 l/min for the Andersen sampler. The particles are deposited on paper or membrane filters, sticky tape, glass or stainless steel surfaces, or agar in Petri dishes.

The trace metal content of filter materials and impaction surface coverings themselves have been investigated extensively because of its possible interference with the particulate analyses. Dams et al.[353] tested

10 filter materials by NAA using lithium-drifted germanium (Ge[Li]) detectors or their spectrometers and found high trace element concentrations in most of the materials. Cellulose paper and membrane filters had the lowest trace metal content, 7-30 ng/cm^2 zinc, compared with filters such as polystyrene, which had 60-515 ng/cm^2 zinc. Bodart *et al.*[137] analyzed finely dispersed filter paper for 10 elements using XRF and lithium-drifted silicon (Si[Li]) and Ge(Li) detectors and found 0.13 $\pm$ 0.04 μg/cm^2 zinc. Birks *et al.*[115] determined 17 elements in membrane and paper filters and found 0.2-30 μg/cm^2 zinc through XRF measurements, compared to Bowman *et al.*, who found 0.007-0.025 μg/cm^2 zinc by NAA.[148a] Birks *et al.* also reported EPA figures based on OES that showed glass fiber filters to have a very high zinc content (160 μg/cm^2). Robertson[1353] summarized contamination problems in trace element analysis and gave the zinc content of polyethylene filters as 25-300 ppb, membrane filters as 2.4 ppm, polystyrene as 4 ppm, paper as 27 ppm, and tissue paper as 49 ppm; he concluded that paper and membrane filters were suitable for collecting particulates from the atmosphere.

Midget impingers also are mentioned as particle collectors in the air pollution literature.[910] They resemble gas scrubbers in that the air stream from a nozzle is bubbled through a liquid in a container. The particles are collected in the liquid by an impaction mechanism although the liquid also has a scrubbing action on the gas stream. Various sizes of impingers are marketed, the midget impinger mentioned above accepting an air flow of 2.8 l/min. These collectors have the disadvantage that some zinc may leach out of the particles into the liquid to cause a negative error, or else some zinc from the container walls may transfer over to the

particulates to give a positive error (contamination). This collection device is popular because of its simplicity and low cost.

When particles are to be collected over an extended period of a month or so and they are large enough (5 μm or larger) to settle out as dust without the aid of motorized blowers, then they may be sedimented into suitable containers using standardized methods such as the Intersociety Committee's tentative method for analyzing dustfall from the atmosphere,[780] or the procedure for collecting and analyzing dustfall proposed by the American Society for Testing and Materials.[30] The advantages of simplicity, low cost, and unattended operation sometimes may be offset by contamination from trees, insects, bird droppings, or curious passersby. Sedimented particles that have settled out on soil and vegetation by the natural action of winds and rain are sometimes studied as alternatives to collection in sedimentation bottles. They have provided valuable information on the amount of zinc and other elements carried in the atmosphere.[198, 765, 891, 1638]

Less frequently used collectors include electrostatic precipitators which impart an electrical charge to the particles so that they are attracted to an oppositely charged electrode. The collector electrode may be covered with a membrane filter for ease in handling the collected material, or with a sample grid for subsequent instrumental analysis, as with an electron microprobe. This type of collector has a high collection efficiency and does not damage the particles physically, but its awkward handling in the field accounts for the poorer incidence of its use. Cyclone samplers which impart a centrifugal motion to the air stream and collect the particles in sharp size fractions above about 5 μm through inertial effects are becoming available and are enjoying increased use. Therman precipitators that impart a thermal

gradient to the air stream and collect particles on the colder of two plates are not used often for particle collection in air pollution studies because of small handling capacities and the extensive cleaning required between samples.

Determination of Zinc in Atmospheric Particulates

Most of the work in this field has concentrated on pollution, with interest directed toward the simultaneous determination of all the metals present in the atmosphere rather than zinc or any other element in particular (except for lead and mercury). Large numbers of low cost, multielement analyses of high sensitivity and good precision and accuracy have been needed most, and this need is being satisfied by the development of sophisticated computer-assisted instrumental methods. Several reviews [564, 815, 758, 1818] are devoted almost entirely to instrumental methods of analysis.

In contrast, classical wet methods of zinc determination as exemplified by the absorption spectrophotometry of dithizone complexes,[428] are slow and expensive. Also, they are suitable for determining only one element at a time and subject to interferences from the other elements present in the particulates. This is not to say that wet chemistry has been discarded; on the contrary, wet chemical manipulations are indispensible for the dissolution of particulates for AAS or polarography, for chemical separations in activation analysis, and for destruction of filter substrates when required. Analytic techniques reported for zinc are NAA, SSMS, XRF spectrometry, OES, AAS, and the classical techniques of absorption spectrophotometry and polarography.

Absorption Spectrophotometry

Although the literature is heavily oriented toward instrumental methods which require a minimum of sample handling (and often not even sample dissolution), this wet-chemistry technique continues to be used because of its modest apparatus requirements, good sensitivity, and potential for high accuracy. In absorption spectrophotometry the particulates are dissolved, interfering elements are removed or sequestered, and the visible or ultraviolet light absorption of a suitable colored complex is measured in solution at a characteristic wavelength.

Marshall et al.[978] described the determination of zinc oxide particulates in air by collection on a membrane filter, dissolution in dilute hydrochloric acid, and colorimetric determination using 4-(2'-thiazolylazo)-resorcinol (TAR) at a wavelength of 530 nm. Zinc oxide concentrations up to 17.4 mg/m^3 were measured, more than twice the threshold limit value set by the United Kingtom. Sereda and Artemova[1471] collected particulates on a filter, dissolved them in dilute hydrochloric acid, separated out interfering metals on an anion exchange column, eluted the zinc with 0.65 M hydrochloric acid, and determined it colorimetrically using sulfarsazen* at 510 nm. The method detected 0.04 µg/ml zinc. Krylova[882] sampled particulates at a rate of 10-15 l/min, dissolved them in 1:1 hydrochloric acid, precipitated interfering metals as dithiocarbamates, and determined zinc colorimetrically using sulfarsazen at a wavelength of 500 nm. The sensitivity was 0.5 µg zinc in 5 ml solution. Further information on absorption spectrophotometry for trace metal determinations has been summarized by Weiss.[1749]

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* Benzene sulfonic acid, 4-[{4-[3-(2-arsono-4-nitrophenyl)-1-triazenyl] phenyl} azo] —, monosodium salt. C. A. Registry No. 1772-02-07.

Atomic Absorption Spectrometry

AAS measures the absorbance of light by a sample when it is present as an atomic vapor at a specific wavelength that is characteristic of the individual element being determined, here 213.9 nm for zinc. A light source often containing the element of interest incorporated in one of the electrodes emits the specific line spectrum of this element, and a simple monochromator isolates the wavelength to be measured. Absorbance is proportional to the concentration of ground state atoms when the vaporized sample is introduced into the light path in the atomic state. The production of vaporized sample in the atomic state usually is accomplished by atomizing a solution of the sample into a flame, or by vaporizing the solution on a tungsten or tantalum ribbon, graphite rod, or in a graphite furnace. The method is rapid, sensitive, and highly specific for metals, although it is subject to some interferences from anions and organic materials. It is well suited to accurate determination of zinc in atmospheric particulates.

Atomic fluorescence spectrometry (AFS) is a related analytic technique in which the optical path is broken at a 90° angle and the stimulated visible and ultraviolet emission from the atoms is observed rather than their absorption. When the direct flame emission is observed without a separate light source, the technique is called flame emission spectrometry (FES). Burrell[206] has reviewed these three techniques.

The analytic procedure of AAS involves collection of particulates on a filter, acid dissolution of the sample, destruction of organic matter including the filter, and measurement against suitable standards. In an extensive review[767] of the use of AAS for determining trace metals including

zinc in atmospheric particulates, Hwang noted a yearly average of about 0.001 $\mu g/m^3$ zinc in urban air samples from 22 cities with incidences up to 1.6 $\mu g/m^3$ The unsuitability of glass fiber filters (160 $\mu g/cm^2$ zinc impurity) and the acceptability of silver and cellulose membrane filter materials (0.01 and 0.002 $\mu g/cm^2$ zinc, respectively) were mentioned.

Thompson et al.[1609] determined zinc and 12 other metals by collection on glass fiber filters and oxidation with a low-temperature asher to destroy organic materials (oxygen passing through a corona discharge formed a strongly oxidizing oxygen plasma which ashed samples at 150 C instead of the 500 C used in conventional furnace ashing). The particulates were dissolved in acid and analyzed by AAS at a wavelength of 213.8 nm. The technique gave a detection limit of 0.2 ng/m^3 for zinc based on a 2,000 m^3 air sample. The sample preparation procedure was stated to be suitable for FES as well, for which a zinc detection limit of 120 ng/m^3 was found.

Ranweiler and Moyers[1316] measured 22 metals in atmospheric particulates by collection on polystyrene filters using high volume samplers, dry ashing at 400-425 C, and dissolution with a mixture of hydrofluoric, hydrochloric, and nitric acids in Teflon bombs. After appropriate dilutions and matrix corrections by adding lanthanum and cesium, zinc and 21 other elements were determined. High zinc concentrations in the reagent blanks were attributed to filter and reagent impurities, and urban air was reported to contain 0.15 $\mu g/m^3$ zinc.

The usual means of producing atomic vapor by nebulizing a sample solution into a flame requires relatively large volumes of solution, a disadvantage in air particulate analyses in which samples are limited. Recently, flameless devices have been introduced to improve sensitivity and decrease sample requirements from milliliters of solution to microliters

of solution. Matoušek and Brodie[167, 986] filtered air particulates directly into perforated graphite cups containing membrane filters. The cup was part of a carbon rod furnace in the instrument light path, and direct determinations of lead and cadmium could be made in air samples as small as 200 ml. Although zinc was not reported, the technique is mentioned because it is novel, and appears to be suitable for the determination of many more metals than lead and cadmium.

The necessity of destroying all organic matter, including membrane, cellulose, and polystyrene filters, has aroused some controversy over suitable procedures for accomplishing this without loss of the volatile zinc. Morgan and Homan, in unpublished work mentioned by Hwang[767] and Thompson et al.,[1609] reported the recovery of only 39% of the zinc after ashing samples in a furnace at 550 C, compared with 96% recovery with a low-temperature oxygen plasma. Dissolving the membrane filter in acetone before ashing in a furnace at 550 C has been recommended.[766,767] Van Raaphorst et al.[1681] summarized the contradictory claims regarding zinc losses during dry ashing and reported his results of radioactive tracer experiments, which showed no volatilization of zinc after dry ashing in porcelain crucibles for 20 h at temperatures to 1,000 C, and no adsorption of zinc on crucible walls to 550 C. Kometani et al.[866] studied dry ashing of airborne particulates on paper and glass fiber filters as a preliminary step in analysis by AAS, and also concluded that losses were not from volatilization but from formation of insoluble silicates. It is inferable from these papers that difficulties encountered with high temperature ashing had a variety of causes and volatilizing of zinc was less important than formation of insoluble slags.

Kleinman et al.[853] and Kneip et al.[861] correlated the results of AAS analysis of atmospheric particulates with meteorologic phenomena. The mixing height of the atmosphere, taken as the height of the inversion layer and measured daily with balloon-borne temperature and altitude sensors, was considered to limit the dispersion of pollutants rising into the air from emission sources at ground level. Multiplying this height in meters by the wind speed in meters/second yielded a product related to the zinc concentration and defined as the dispersion factor, with units of m^2/sec. Marked seasonal variations for zinc and for the total particulates, involving spring minimums and summer maximums, were attributed to changes in the atmospheric capacity for mixing, as indexed by this dispersion factor. For additional information on applications of AAS, attention is invited to Price's work.[1300]

Optical Emission Spectroscopy

Optical emission methods measure the wavelength and intensity of visible and ultraviolet radiation emitted from excited electronic states of atoms introduced into arc and spark excitation sources. They are suitable for rapid, semiquantitative, multielement surveys of airborne particulates as well as for accurate quantitative measurements of individual elements. An emission spectrographic laboratory utilizes critically aligned, vibration-free optical and excitation instruments, computer-based data processing, and personnel with a high degree of skill and training. Nevertheless, the speed, high sensitivity, and broad applicability of OES make it one of the most popular analytic techniques for determining zinc in the atmosphere.[1390]

The technique generally involves powdering or ashing the samples, mixing them with a buffer and an internal standard, and arcing in graphite electrodes. Lander et al.[894] described the spectrographic determination of zinc and 11 other elements in airborne particulates. Samples collected on paper or membrane filters were analyzed without further treatment by rolling the papers tightly into cylindric graphite sample electrodes and burning them with high voltage spark excitation in an oxygen atmosphere. Zinc was measured at a wave length of 213.9 nm. Calculations and tabulation of results were performed by a computer. Individual filters 25 mm in diameter, sampled at a rate of 14 1/min, contained up to 1.5 µg zinc, with the detection limit judged to be 0.1 µg zinc. Coefficients of variation (the measure of precision) ranged from ±9 to ± 60% for the individual elements.

Sugimae[1562] collected airborne particulates on a silver membrane filter, dissolved the filter in dilute nitric acid, precipitated the silver with hydrochloric acid, evaporated the solution to dryness and determined zinc and 11 other elements simultaneously by emission spectrography using dc arc excitation. Zinc was determined in the concentration range 0.13-2.5 µg/m^3, based on a sampled air volume of 400 m^3 (50-1,000 µg/filter) with a precision of ± 15%, using the analytic line pair of zinc I 334.5020 nm and indium I 303.9356 nm.

Imae et al.[778] collected particulates on membrane filters for 10 days using a low volume sampler with a flow rate of 20 1/min, burned the filters with ethanol (C_2H_5OH) in a quartz boat and further ignited the carbonized ash in a low temperature plasma to remove the carbon and residual organic materials. They mixed the ash with indium and palladium oxides, buffered with lithium carbonate

and graphite, and excited the sample in an ac arc to determine zinc and 12 other trace elements. The urban zinc concentration was reported as 0.14-1.02 $\mu g/m^3$ with a precision of $\pm$ 16.5%, using the 328.23 nm analytic line for zinc. Metallic impurities in hydrochloric acid extracts of 105 mm diameter filter materials also were measured: zinc content was 0.4-1.0 µg for 5 membrane filters, 10.0 µg for 1 polystyrene filter, and 73 µg-10.3 mg for 6 glass filters.

Seeley and Skogerboe[1466] avoided problems of filter contamination and disposal by using the graphite spectroscope electrodes as filters for atmospheric particulates, sampling at a rate of 1 l/min. They determined zinc and 13 other elements directly by emission spectroscopy using dc arc excitation with indium as an internal standard. Their detection limit for zinc was 3 ng when the analytic wavelength of 334.502 nm was used.

Lee et al.[914] reporting on the results of emission spectrographic analyses of particulates collected by the National Air Surveillance Cascade Impactor Network (NASN), emphasized the variation of metal concentrations with particle size. Particulate matter collected biweekly over 24-h periods with a 5-stage Andersen sampler was extracted with nitric acid, buffered with lithium chloride, and analyzed on a 2-meter spectrograph employing a rotating disc and high-voltage spark source. Indium and yttrium were taken as internal standards. The annual concentration levels observed for zinc were 0.1-1.7 $\mu g/m^3$, which were intermediate between highs for iron and lead and lows for nickel and vanadium. Particle size distributions were correlated with metal concentrations for zinc and 11 other elements. About 70-80% of the zinc particles were smaller than 2 µm, and 40% were smaller than 1 µm. No seasonal pattern was discerned, but a geographic pattern hinted at possible urban sources of zinc in the atmosphere.

X-Ray Fluorescence Spectrometry

XRF spectrometry, also known as X-ray emission spectrometry, measures the characteristic photon or X-ray emissions that result when higher energy electrons fall into a K or L shell whose electron was previously ejected by irradiation with high energy photons or particles. The energy of the emitted X radiation is characteristic of the elements present in the sample, and the frequency with which the transitions occur is proportional to the quantity of element present. All elements above sodium can be determined rapidly without any sample preparation.

The instrumentation required includes an excitation source which may be an X-ray tube, radioisotope, or other source of electrons, protons, or ions; a sample holder; an analyzer to resolve the emitted X rays into an energy spectrum by wavelength dispersion (such as crystal diffraction, or bandpass filters) or by energy dispersion; and an X-ray detector. Detectors used with wavelength dispersion spectrometers may be scintillation detectors, or gas-filled or gas-flow proportional detectors. Detectors used with energy-dispersive spectrometers are the solid-state Si(Li) detectors, with an output pulse height proportional to the incident energy. A great many combinations of source, analyzer, and detector are possible.

Several very readable introductions to the principles of XRF analysis exist.[114, 115, 531] Four types of excitation (X-ray tubes, fluorescers, radioisotopes, and high energy ions) are compared, as well as two types of data analyzers (crystal spectrometers and energy-dispersive detectors) for determining 10-20 elements including zinc in air particulates. A multichannel crystal spectrometer instrument is recommended as the optimal technique for measuring at least 12-14 elements simultaneously. If only

one or a few elements are of interest, then energy dispersion analysis with a low power X-ray tube or high activity radioisotope and a simple proportional counter may be an adequate alternative to sophisticated laboratory instrumentation. Zinc detection limits for 100-sec measurements of prepared standards ranged from 51 ng/cm^2 for X-ray tube excitation and wavelength dispersion to 1,400 ng/cm^2 for isotope (iron-55) excitation and energy dispersion. XRF results for zinc and 16 other elements were comparable to atomic absorption measurements made on the same particulate samples. Studies of elemental impurities in filter materials have been mentioned earlier.[115]

Rhodes et al.[1337] compared energy-dispersive XRF spectrometry using radioisotope sources with AAS for determining zinc and 16 other elements in particulates collected with high volume samplers on cellulose filter paper. Air pollution data presented for urban areas averaged 0.13 μg/m^3 zinc, compared with a national average of 0.67 μg/m^3. The X-ray analyses produced consistently lower results than the atomic absorption determinations, and this discrepancy was thought to be an effect of particle size of the samples.

Luke et al.[958] compared XRF spectrometry with AAS for analyzing particulates collected on filter paper. Zinc and six other elements were determined by direct X-ray analysis, X-ray analysis of samples after wet chemical separations, and AAS. The results, between 2-100 μg/cm^2 for zinc, were comparable for all 3 techniques.

Leroux and Mahmud[924] analyzed particulates collected in an urban area on membrane filters and found 0.08-4.03 μg/m^3 zinc. The analyses took only 5 min/element. Giauque et al.[529] used X-ray-induced XRF spectrometry to determine zinc and other elements in particles collected over

2-h intervals for 24 h with a 4-stage impactor. Detection limits for zinc were stated to be 4 ng/m^3 on membrane filters and 2 ng/m^3 on Mylar filters. Diurnal charts showing changes of zinc concentrations with time were presented for several cities. Cares[230] described X-ray spectrometric procedures for airborne dusts and included a detailed method for preparing reference standards, but zinc was mentioned only in passing. Dittrich and Cothern[386] analyzed trace metals in urban atmospheric particulates using a high volume air sampler to collect particles on filter paper. A γ-ray source (americium-241) and a bremsstrahlung excitation source (promethium-147) both were used to produce X rays. Data for zinc and six other elements were presented as counts rather than as μg/m^3. Mitsugi et al.[1071] collected air particulates on filter paper using a high volume sampler, and determined zinc and lead concentrations directly; also, samples collected with an electrostatic precipitator were briquetted with boric acid and analyzed. Zinc concentrations in the range of 0.25-1.06 μg/m^3 agreed with results yielded by AAS. Bodart et al.[137] analyzed particulates on filter papers by irradiating the filters with 1.7 million electron volt (MeV) protons from a low energy Van de Graaff generator, and detecting XRF with Si(Li) and Ge(Li) detectors. Standards were prepared from pure salts and powdered paper filters. Findings were presented for 10 elements: results for zinc fell in the 1.40 $\pm$ 0.14 μg/cm^2 range.

Mizohata and Mamuro[1073, 1074] used americium-241 excited, energy-dispersive XRF spectrometry to determine zinc and 11 other elements in airborne urban dust samples. Corrections for sample loading on membrane filters were made by comparing the X-ray data with determinations from instrumental NAA. (The measured X-ray emission of zinc decreased about 20% as the mass loading of the filter increased from 0.2 to 7.5 mg/cm^2 because of self-

absorption in the sample.) Actual dust samples collected with a low volume air sampler were found to contain 1.1-48.7 $\mu g/cm^2$ zinc, as measured by the two techniques.

Hammerle et al.[625] compared XRF spectrometry with NAA for determining 10 elements in atmospheric particles. The zinc analyses for both techniques agreed within 6.7% of the average of 0.28 $\mu g/m^3$ found in urban particulates.

Cooper[314] compared particle- and photon-excited XRF determinations of zinc and about 20 other elements in urban aerosols collected on filter paper, and in rural aerosols collected on Mylar film in an impactor. The samples were cemented as thin samples or briquetted as thick samples, and analyzed by excitation with either high energy α-particles (80 MeV and 30 MeV) or protons (4 MeV and 2 MeV). Cooper also studied photon excitation with a molybdenum target X-ray tube, and with high intensity radioisotope excitation using iron-55 and cadmium-109 sources, and measured the energy spectrum with a Si(Li) detector. Zinc was reported at a concentration of 1-5.6 ng/m^3 in typical urban particulates. Photon excitation methods were concluded to be significantly more sensitive than high energy α-excitation, with the additional advantages of small size, easy portability, and applicability to a wide variety of samples for trace element analysis of environmental particulates.

This brief survey of the application of XRF spectrometry to the determination of zinc in air particulates shows that it is a popular analytic technique because 20 or so elements may be determined simultaneously and directly on filters without prior sample manipulations such as acid dissolution, extraction, or ashing. What cannot be emphasized strongly

enough is the necessity of having reliable calibration standards because the technique is substrate-sensitive. Dried solutions of pure salts on filters may not simulate collected particulates closely enough, because the latter are mixtures of many different inorganic and probably organic materials with different particle sizes and shapes. It seems reasonable to use a uniformly deposited, exhaustively analyzed natural material for an X-ray calibration standard to supplement the data obtained with pure salts.

Neutron Activation Analysis

Activation analysis is similar to XRF or emission spectrometry except that it is the atomic nucleus rather than an extranuclear electron which is energized by irradiation with energetic particles or photons. The emitted radiation of the nucleus as it decays radioactively is measured. The energy and type of the emitted radiation as well as its rate of decay identify the nucleus, and the intensity of the radiation is a measure of the amount of element present. The technique generally involves irradiation with thermal neutrons, chemical separation of the activated species, and measurement of the emitted radiation, preferably with solid-state detectors coupled with multichannel analyzers and computer-controlled data processing to convert spectral counts and energies to elemental concentrations. Although a variety of neutron sources is available (neutron generators and isotopic neutron sources such as californium-252), the trace metal concentrations in airborne particulates are so low that high thermal neutron fluxes of the order of 10^{12} or 10^{13} n/sec/cm^2 are required and these fluxes are available only from nuclear reactors. Chemical separations of activated species may be required if the energies of emitted radiation from many elements overlap;

however, the high resolutions available from the new solid-state Ge(Li) γ-ray detectors make direct sample assays possible without the need for wet chemistry. Two reviews[815, 835] give a concise introduction to the subject.

Dams et al.[352, 354] and Rahn et al.[1311] have described the determination of 33 elements in air pollution particulates directly on filters without prior chemical separations. Particulates collected on polystyrene filters were irradiated for 2-5 h (for long-lived isotopes such as zinc) in a nuclear reactor at a thermal neutron flux of 1.5×10^{13} n/sec/cm^2 together with a standard mixture of elements in a polyethylene bottle. After irradiation, the samples and standards were counted with a Ge(Li) detector coupled to a 4,096 channel pulse height analyzer. The spectra were recorded on seven-track magnetic tape for subsequent data reduction by computer. Zinc was determined as the 13.8-h zinc-69 metastable radioisotope, using the 438.7 kiloelectron volt (KeV) γ-ray radiation. The detection limit for zinc was 0.2 μg, which corresponded to an urban air concentration of 0.02 μg/m^3 for a 24-h sample. The authors commented that emission spectrography would detect about 0.24 μg/m^3 zinc in such a sample. Suspended particulates contained 180-1,690 ng/m^3 zinc (standard deviation, 10-300 ng/m^3). Dams and coworkers[353] used this analytic technique to evaluate filter materials and study airborne trace element distributions.[640, 1311] Zinc and antimony exhibited similar well-defined distribution patterns which were correlated to geographic areas.

Kuykendall et al.[887] used automated instrumental NAA to determine zinc and 39 other elements in air filter samples. Particulates collected on filter paper with a high volume sampler operating for 24 h at a flow rate of 1,223-1,631 m^3/day were irradiated in a neutron flux of 5×10^{12} n/sec/cm^2 for 8 h (for long-lived radioisotopes such as zinc), and counted

directly with a Ge(Li) detector coupled to a 3,200 channel pulse height analyzer with computer-compatible magnetic tape readout, sample changer, and system controller which permitted continuous unattended counting of many samples. Zinc was reported in the range of 48-1,453 ng/m^3, with a mean of 410.5 ng/m^3, which was well above the stated detection limit of 120 ng/m^3.

Schramel et al.[1440] determined zinc and 11 other elements in dustfall samples collected for 1 mo periods in polyethylene bottles, thereby avoiding the limitations of filter blanks. The particulates were sealed in quartz ampoules and irradiated for 24 h in a thermal neutron flux of 10^{13} n/sec/cm^2. They were cooled for 3 days, and the zinc group was chemically separated from the other elements and counted with a Ge(Li) detector connected to a 4,096 channel pulse height analyzer. High zinc concentrations were noted in areas of heavy vehicular traffic (16.26 mg/g), which were thought to be caused by airborne rubber particles from automobile tire wear.

Winchester[1788] discussed the significance of the results of NAA as they related to occurrence and transport of pollution and natural substances in the air. He recommended that sampling from aircraft be seriously considered for acquisition of improved data.

Particulates have been collected with excellent size fractionation on polyethylene films in a cascade impactor[49, 539, 548] and analyzed for 18 elements by instrumental NAA. Zinc occurred predominantly in the finest particles, in 100 times the concentrations found in the coarsest particles. Actual particle sizes were not determined. Statistical correlations with elemental abundances from known sources such as the earth's crust, marine aerosols, coal and oil combustion, and automobile

exhausts did not explain the zinc enrichment in the atmosphere. The fact that the greatest amounts of zinc were found in the smallest particles seemed to suggest vaporization followed by condensation, that is, a combustion source. It was suggested[539] that tire dust generated by abrasion would most likely consist of large particles, so that the hypothesis of tire wear as a source of zinc[891, 1440] would not be consistent with measured particle size distributions.

Pillay and Thomas[1247] described the analysis of airborne particulates using sequential air sampling onto special high-purity filters, high flux (3×10^{13} n/sec/cm^2) thermal neutron irradiation, and high-resolution γ-ray spectrometry to determine zinc and 18 other elements. Instrumental neutron activation for multielement analysis of airborne particles has been described by other investigators.[252, 351, 547, 556, 795]

Aras et al.[49] determined 14 elements including zinc by an instrumental photon activation technique. Atmospheric particulates were irradiated with bremsstrahlung from 35 mev electrons produced with a linear accelerator, and the emitted γ-ray radiation was counted with a Ge(Li) detector. Zinc was determined by the zinc-68 (γ,p) copper-67 reaction, and was reported in the range of 0.09-0.22 μg/m^3 at several sampling locations. The detection limit for zinc was reported to be 3 ng/m^3, compared with 4×10^{-5} ng/m^3 for instrumental NAA, and 0.0025 ng/m^3 for flame methods. Therefore, it was concluded that instrumental photon activation analysis was a useful technique for analyzing particulates. Instrumental NAA is the method of choice for many investigators who determine zinc and other elements simultaneously in air particulates. The wide applicability of this analytic technique has been thoroughly demonstrated.[379]

Mass Spectrometry

Mass spectrometry of airborne particulates is usually taken to mean SSMS, in which samples introduced as electrodes in an evacuated chamber are pulsed with a 1 megahertz (MHz) radio frequency (rf) voltage of 20-80 keV to yield ions of all the elements present. The ions are accelerated with a 24-kV dc potential, focused in an electrostatic sector, and dispersed according to mass:charge ratio in a magnetic sector for photographic or electronic detection and measurement. The technique is highly sensitive: it can detect all the elements simultaneously including adsorbed gases. Furthermore, the spectrum is simple (consisting mainly of singly-charged positive ions), and sample preparation is minimal, requiring only 10-100 mg of solids for analysis. Some limitations include spark-source instability leading to nonuniform ion production, and the relatively poor sensitivity and reproducibility of photographic plates used for detection and measurement.

Brown *et al.*[172] and Brown and Vossen[176] reviewed research on solid-state mass spectrometry and presented analyses of particulates on membrane filters. Table 13-1 lists 46 elements including zinc, which were determined in 2.65 mg particulates collected on a membrane filter.[172]

TABLE 13-1

Analysis of Particulate Matter from Mine Interior for Trace Elements[a]

Element	Concentration[b]	Element	Concentration[b]
Uranium	0.006	Gallium	0.18
Bismuth	0.02	Zinc	1.1
Lead	0.07	Copper	2.3
Samarium	0.01	Nickel	2.0
Neodymium	0.12	Cobalt	0.03
Praseodymium	0.03	Iron	210.0
Cerium	0.12	Manganese	0.12
Lanthanum	0.06	Chromium	0.45
Barium	1.2	Vanadium	0.44
Iodine	0.01	Titanium	42.0
Tellurium	0.01	Scandium	0.08
Cadmium	0.01	Calcium	35.0
Silver	0.02	Potassium	44.0
Molybdenum	0.04	Chlorine	0.61
Niobium	0.02	Sulfur	8.3
Zirconium	0.16	Phosphorus	0.81
Yttrium	0.02	Silicon	780.0
Strontium	0.77	Aluminum	750.0
Rubidium	0.02	Magnesium	2.1
Bromine	0.03	Sodium	2.0
Selenium	0.02	Fluorine	0.005
Arsenic	0.07	Boron	0.01
Germanium	0.05	Lithium	0.02

[a]Data from Brown et al.[172]

[b]Weight retained on filter in μg; total sample weight 2.65 mg on Millipore filter.

In an interesting variation, Perry[1233] mentioned that trace metals on filters might be converted to volatile metal oxinates or β-diketonates, extracted into an organic solvent, and identified and determined quantitatively using conventional high-resolution mass spectrometers of the types used to analyze organic materials. Standardization with pure metal alkyls or chelates would be required. Only findings for lead tetraethyl were presented. The use of such metal-organic complexes has been proposed for gas chromatographic analysis.[917] Since zinc forms volatile chelates with a variety of β-diketones such as 2,4-pentanedione, hexafluoro-2,4-pentanedione, 2,2,6,6-tetramethyl-3,5-heptanedione, and 2-thio-2,4-pentanedione,[917] it may be possible to determine zinc by mass spectrometry. Such determinations apparently have not been reported yet.

Despite its broad capabilities, SSMS does not appear to have been used extensively to analyze airborne particulates.

Voltammetry

In common with other techniques requiring sample dissolution, buffering, and other wet-chemical processing, electroanalytic methods are not described in the air pollution literature as frequently as multielement techniques that require few sample manipulations. With such attractive features as high sensitivity and selectivity, low cost, and a modest multielement capability, voltammetry should be used more extensively in environmental analysis than it is.

In voltammetry (polarography), current-voltage relationships measured during electrolysis are used to identify and quantitate the ions reacting

at one of the electrodes (generally a dropping mercury electrode). Many variations of the basic technique have been developed, and instruments are available for conventional dc polarography, derivative dc polarography, cathode ray polarography, ASV, and ac, square wave, and pulse polarography. The various names refer to the types of voltage applied to the electrodes and the forms of data presentation. The highest sensitivity is afforded by ASV, in which the elements are plated from solution onto a cathodic electrode and then oxidized into solution by applying a voltage scan in the anodic direction. The peak currents arising from the oxidation of the ions of interest are measured to yield results down to nanogram and picogram concentrations in solution.

Landry[895] described early work on polarographic zinc determinations in the atmosphere. More recently, ASV has been used[307] to determine trace amounts of zinc in airborne particulates: particles were collected on a membrane filter, which was ashed to destroy the filter and organic materials; the residue was digested in nitric and hydrofluoric acids. The solution was diluted and analyzed by stripping analysis, using 0.1 M potassium nitrate as supporting electrolyte, by plating at -1.2 V for 3 min, then scanning from -1.2 V to + 0.1 V against a standard calomel electrode. Results for zinc were 0.020-0.93 $\mu g/m^3$; they were compared with atomic absorption analyses carried out on the same samples. Matson et al.[989] used ASV to determine zinc in tissue, blood, urine, and hair below the nanogram level and found excellent agreement with results from AAS and NAA. Their technique employed perchloric acid digestion, addition of 0.5 M salt (NaCl), 1 M sodium acetate, and 0.5 M ethylenediamine, plating for 30 min at -1.3 V, and then stripping

at 66 mV/sec to a potential of +0.1 V against a standard calomel electrode. The method should be suitable for analysis of particulates.

To sum up, the variety of analytic methods and sampling procedures that have been used complicates comparing results from different investigators that might allow the testing of theories about zinc sources, atmospheric transformations, or removal mechanisms. Katz[823] has suggested the initiation of collaborative testing programs to evaluate the various analytic methods for specificity, selectivity, sensitivity, range, precision, and accuracy; however, very often these programs evaluate the capability of the operators rather than the analytic methods used. Von Lehmden et al.[1705] attempted to evaluate methods of analysis for coal, fly ash, fuel oil, and gasoline: 9 laboratories determined concentrations of zinc and 27 other elements in the same samples using SSMS, OES, INAA, AAS and ASV. Table 13-2 presents interlaboratory results obtained for these methods of analyzing zinc. Because the wide variations reported are a consequence of differences in sample handling and instrument operating procedure, results are difficult if not impossible to interpret. Comparisons of data from different laboratories (for example, the different literature cited in this chapter), could lead to erroneous conclusions.

TABLE 13-2

Collaborative Test Results for Zinc Determinations in Various Materials[a]

Laboratory	Method of Analysis	Coal, ppm	Fly Ash, ppm	Fuel Oil, ppm	Premium Gasoline, µg/ml	Low-Lead Gasoline µg/ml
1	SSMS[b]	100	200	0.5	0.2	-
3	SSMS	5	1,000	-	1.0	4.0
6	SSMS	6.6	330	-	-	-
1	OES[c]	100	100	0.4	0.2	-
3	OES	50	200	2.0	-	-
2	INAA[d]	-	-	1.4	0.36	0.43
3	INAA	-	-	1.3	-	-
4	INAA	100	-	-	3.0	-
5	INAA	-	-	-	-	-
8	INAA	-	-	-	-	-
3	AAS[e]	-	600	-	-	-
9	DRES[f]	-	350	-	-	-
1	AAS	-	-	2.0	1.0	-
7	ASV[g]	-	-	-	0.12	0.096

[a] Derived from von Lehmden et al.[1705]
[b] SSMS - spark-source mass spectrometry
[c] OES - optical emission spectrography
[d] INAA - instrumental neutron activation analysis
[e] AAS - atomic absorption spectrometry
[f] DRES - dissolution followed by dc arc excitation in emission spectroscopy
[g] ASV - anodic stripping voltammetry

CHAPTER 14

SUMMARY

PROPERTIES AND USES OF ZINC

As the fourth most widely used industrial metal, zinc serves the consumer in a form not easily recognized in the end product. It is a corrosion protection for steel (galvanizing); an alloying metal with aluminum, magnesium, and titanium in mass-produced precision parts (die-casting); an alloy with copper for brasses; a chemical for white paint pigment; and a vital ingredient in compounding rubber products. Zinc compounds have long been used pharmaceutically, although some newer applications in physiology and medicine are still being evaluated.

NATURAL SOURCES AND DISTRIBUTION OF ZINC

Zinc is found in most soils, but some areas are deficient in it. Differences of zinc in soil and water can influence the zinc content of plants and animals found in these areas and in the products derived from them.

MAN-MADE SOURCES OF ZINC

Metallurgic operations, primarily mining and smelting of lead and zinc ores, contribute appreciable zinc contamination to air, water, and soil. However, this contamination is normally limited to areas near point sources. Few data are available on airborne concentrations of zinc near metallurgic operations. However, evidence of elevated soil and vegetation zinc concentrations near these operations implies that airborne levels of zinc in these areas are or were appreciably elevated. The closing down of many older primary zinc operations has reduced environmental contamination and presumably new operations will have better particulate controls.

Available data, although scanty, indicate that airborne zinc concentrations in the United States away from point sources are generally low (less than 1 $\mu g/m^3$). Zinc in drinking water in the United States rarely exceeds the drinking water standard of 5 mg/l. Data are conflicting on the contribution of mobile sources to zinc contamination near roadways. If mobile sources do contribute markedly to the zinc roadside dust, the zinc probably is generated from zinc compounds in tires and motor oil. Sewage sludges also may contain high concentrations of zinc and other metals. Such sludges may not be suitable for indiscriminate use on agricultural land.

Few data are available on the zinc content of solid wastes and the contribution such waste may make to total environmental zinc levels. Information that exists on the quantity of zinc-65 released into waters from nuclear power plants would seem to indicate that levels are within acceptable limits.

ZINC IN PLANTS

Aquatic Plants

Trace amounts of zinc are essential for normal growth and development of aquatic plants. Uptake of zinc is accomplished by ion exchange and metabolic assimilation processes. Aquatic plants can accumulate much more zinc by sorption than is needed for metabolic activity. Rooted plants may take up zinc from sediments as well as from the ambient water, but zinc is usually accumulated by the plant in relative proportion to zinc in the ambient water. Zinc concentration factors in plants as compared to the ambient water solution may range from a few orders of magnitude to 19,000. Therefore, zinc concentration of aquatic plants can range from a fraction of a ppm to several thousand ppm. Zinc concentrations in aquatic plants vary seasonally, and the variation is probably caused by differences in availability and content of zinc in ambient waters as well as factors such as growth rate, supply of other nutrients, and temperature.

Excessive levels of zinc in waters can bring on zinc toxicoses in aquatic plant communities. Tissue sampling and analysis techniques are being developed to establish threshold values for zinc toxicity to aquatic plants. The same analytic data may be used to determine the overall zinc nutritional status in aquatic plants. A primary function of zinc in aquatic plants is as a cofactor for several metalloenzymes and possibly as a stabilizer of the integrity of plant ribosomes. Aquatic plants (mainly algae) have been used extensively to elucidate the metabolic functions of zinc in plants.

Terrestrial Plants

The recognition of zinc deficiency and an understanding of essential functions of zinc in terrestrial plants have been commonplace since 1900, and zinc deficiency is now the most common micronutrient deficiency in the United States. Plants vary widely in sensitivity to zinc deficiency, toxicosis, and tolerance. Climatic and soil factors affecting zinc availability to plants are: amount of soil reserves; soil pH; extensiveness of root zones; microbial and soil organic matter content; soil temperature and moisture; and interactions with other elements. Most soil zinc moves to plant roots by diffusion--convection plays a minor role. Soil intensity and capacity as well as availability of natural and synthetic chelating agents influence the movement of zinc to plant roots in soil. Zinc is taken up by plants as the divalent cation, and both active and passive uptake mechanisms are present. The active metabolic component is most important for continued zinc uptake by plants. Zinc is translocated in the xylem of plants as the divalent cation or as a weak metal-organic complex. Some zinc is redistributed from older to newer tissues through the phloem. Evidence exists for considerable interaction of zinc with phosphorus, iron, copper, and other elements in the plant. These interactions influence the rate and degree of zinc translocation. Zinc deficiency is often observed in plants that contain less than 20 ppm zinc; the normal plant concentration is 25-150 ppm zinc. Toxicosis

often occurs at concentrations above 400 ppm zinc. Zinc deficiency can be overcome by foliar or soil fertilizer applications of soluble zinc salts. Zinc is intermediate in toxicity to plants among the heavy metals. Several species of plants that are extremely tolerant of high zinc concentrations in soil have been identified and provide an opportunity to establish vegetation on zinc-contaminated soils. The best defined role for zinc in plants is as an enzyme cofactor in various metalloenzymes. The greatest amount of research has been conducted on carbonic anhydrase, but the necessity of zinc to several metalloenzymes is well established. There is also evidence that zinc may function in stabilizing ribosomes.

ZINC IN AQUATIC ANIMALS

Although zinc is ubiquitous in aquatic organisms, the environmental and physiologic mechanisms controlling the biologic availability and accumulation of zinc in tissues are not well understood. In fresh fish muscle, zinc normally occurs in a concentration range of 3-30 ppm. Similar concentrations are observed in muscle tissue of many crustaceans and mollusks, particularly in those from open ocean waters. Organisms from coastal estuarine waters, however, tend to exhibit higher and more variable concentrations of zinc. Oysters have a particularly high affinity for zinc, occasionally accumulating the element to more than 1,000 ppm. The highest concentrations in oysters are usually found in specimens from environments low in salinity. In mussels, scallops, and freshwater bivalve mollusks, zinc is strongly localized in certain organs, whereas in oysters the metal is rather uniformly distributed throughout all tissues. Fish, decapod crustaceans, and certain species of polychaete worms appear able to regulate tissue concentrations of zinc.

Since aquatic organisms spend their entire lives immersed in a complex chemical milieu, it is difficult to distinguish dietary sources, integumentary exchange processes, and excretory and secretory mechanisms. An understanding of the

physicochemical behavior of zinc in the aquatic environment is therefore a requisite for metabolic studies of aquatic organisms. Numerous studies have suggested that various chemical forms of zinc (including organic complexes) occur in natural waters, but their role in biologic accumulation and metabolism of zinc in organisms has not yet been defined.

The disposal of industrial and municipal wastes in the aquatic environment has made zinc toxicosis a potential problem for aquatic organisms. Short-term acute toxicosis experiments have indicated that the median lethal concentration (LC_{50}) for some species may be in the range of 100-300 µg zinc/l. In fish, acute metal toxicosis has been attributed to precipitation of mucus on gills or to other cytologic damage to gill tissue. Systematic studies of the effects of environmental variation on toxic or sublethal effects of zinc have not been performed for most aquatic organisms.

ZINC IN HUMANS

Zinc is an essential element found in every human tissue and tissue fluid. Among the /transition elements, the concentration of zinc in the body is second/ trace only to iron. Approximately /90% of total body zinc is found in muscle and bone, but the highest concentration of zinc is found in endocrine glands, particularly the gonadal system, and in sensory receptors, particularly the retina of the eye. Only a small amount of total body zinc is carried in the blood, with the active fractions bound primarily to either albumin or amino acids, particularly histidine. Zinc is normally excreted in the urine bound to amino acids, but only a small fraction of the circulating zinc is excreted daily. Most dietary zinc is excreted in the feces as a result of lack of absorption or because of resecretion from bile.

Zinc is a critical constituent of DNA polymerase. Without it, protein synthesis does not proceed normally and cell division appears to be abnormal. Zinc deficiency in humans is commonly associated with abnormalities

in those systems in which rapid cell division occurs; hence, growth retardation, hypogonadism, and abnormalities of the gastro-intestinal tract are common complaints. Zinc also appears to be particularly important for growth and development in utero and in early life.

The specific manner by which zinc is transported across the gut is not known, but the process may involve the formation of a low molecular weight organic zinc chelate which moves across the gut primarily in several areas of the small bowel. Studies with zinc-65 suggest that about 65% of this isotope is absorbed by normal humans; however, there is an extremely broad range of absorption of this isotope, a phenomenon quite different from the absorptive characteristics of other trace metals. Zinc malabsorption, which occurs in several gastro-intestinal diseases, is associated with absorption of less than 30% of the zinc-65 presented orally.

Although the specific functions of zinc in various organ systems are unclear, most systems appear dependent upon its presence. In the endocrine system, zinc is associated with the release of several pituitary hormones, the prolongation of action of adrenocorticotropin and insulin and an effect on prostate and testes that appears important in spermatogenesis. Many hormones influence zinc metabolism, and the bases of these changes are related to effects not fully understood.

Zinc is involved in muscle function. Muscle represents the largest single body pool of zinc and the metal may be associated with potentiation of contractibility. Zinc is associated with key enzymes involved with the function of several organ systems, including liver and retina, and with vitamin and hematopoetic function. The association of zinc with alkaline phosphatase and the prominent role of this enzyme in the function of receptor membranes suggests a ubiquitous role for zinc.

In sensory systems, the enzyme needed to convert retinol to retinal in the retina is zinc-dependent; hence visual processes are, in part, dependent upon this metal. In saliva, the zinc protein, gustin, may be important in the growth and differentiation of taste buds. With the isolation of taste bud receptor membranes from the cow, the zinc-dependent enzyme alkaline phosphatase appears to be the most highly concentrated enzyme; demonstration of specific binding of sugars and other tastants to the isolated taste bud membrane are zinc-dependent. Zinc depletion in humans or animals is almost uniformly associated with anorexia and taste loss, conditions usually reversed following administration of zinc.

Although zinc is the fourth most prevalent cation in all brain tissue, little is known of its function. Its location in the cerebellum and in the limbic system and the production of cerebellar dysfunction and mental dysfunction with acute zinc depletion in humans are well established. Severe mental aberrations in acutely zinc-depleted human subjects and poor performance in learning situations by animals made zinc-deficient suggest that zinc is associated with higher brain function as well as several neurophysiologic functions.

ZINC IN THE DIET

Zinc Deficiency

Feeding animals diets low in zinc has decreased growth in all species tested. Other signs of zinc deficiency frequently observed in young animals are parakeratosis, hyperkeratinization, and impaired testicular development. Animals in less rapid stages of growth may have no signs of deficiency other than poor appetite and decreased growth. Wound healing has been impaired in adult animals on a low zinc diet; and reproduction, at least in the female rat, has been severely affected. Since food consumption is consistently reduced in zinc deficiency, some of the problems apparently caused by a low zinc intake are a secondary effect of decreased food consumption.

Sources of Zinc and Amounts Required in the Diet

Zinc in the diet primarily comes from protein. Meat, legumes, and whole grains are good sources of zinc. Zinc intakes considered adequate for various animals range from 20-50 mg/kg diet. Factors other than the actual content of zinc in the diet affect its adequacy, however, since deficiencies have developed in animals eating diets with presumably adequate amounts of zinc.

The recommended dietary allowance (RDA) for adult men and women is 15 mg/day. Low protein diets are apt to have significantly less zinc than this. A higher intake is recommended during pregnancy and lactation.

Since zinc deficiency in pregnant rats has caused congenital malformations, concern has been expressed that congenital malformations in humans might be a consequence of zinc deficiency. Malformations have only been obtained with diets extremely low in zinc and so far only in rats. Whether the results with rats would be obtained in other species is presently unknown.

Interrelationships with Other Components of the Diet

Phytic acid was one of the first constituents of plants to be linked to the decreased availability of zinc from plant sources. Other factors in plants affect zinc availability, but there is little information on them.

High calcium levels in the diet aggravate symptoms of zinc deficiency, particularly in pigs; calcium does not have much effect, however, unless most of the protein in the diet is derived from plant sources, particularly seeds.

Although interrelationships between zinc and vitamins such as biotin and minerals such as cobalt have been suggested, no vitamin or mineral additions have alleviated the symptoms of zinc deficiency. Additional histidine or histamine has alleviated the swollen hock condition that develops in zinc-

deficient chicks, although other symptoms of zinc deficiency in chicks were not affected.

Occurrence of Zinc Deficiency

Severe zinc deficiency caused by low zinc intake is rare. The incidence of marginal deficiency states is largely unknown because of the lack of a good method for assessing zinc status. Low hair zinc levels reported in some American school children suggest that marginal zinc deficiencies do occur. Apparent zinc deficiencies have also been reported in animals fed diets appearing to be adequate in zinc.

Assessment of Zinc Deficiency

Because the chief effects of low zinc intake -- poor appetite and slow growth -- may arise from many causes, a specific test is needed to diagnose zinc deficiency. Plasma and hair zinc levels have been used most frequently to assess the zinc status of humans and animals. Plasma zinc is limited in that it is affected by many things other than zinc intake. Hair zinc reflects zinc status only over fairly long times. Although both measures probably will be low in cases of severe deficiency, they may not be lowered in states of marginal deficiency.

Metabolic Lesions in Zinc Deficiency

Zinc deficiency results in decreased growth of most body tissues, although the effect does not seem to interfere with DNA synthesis *per se*. Indeed, mitotic activity increases in cells in the esophagus and buccal mucosa. Why mitotic activity should increase in a few tissues and decrease in most others is not known. Disturbances in sulfur metabolism, glucose metabolism, bone growth, and reproduction have also been reported. However, the extent to which these differences are caused by altered food intake or reduced growth of the deficient animal rather than the lack of zinc *per se* is unclear.

Zinc in Wound Healing in Animals

Although zinc has been implicated in more rapid healing of wounds, studies with animals indicate that additional zinc is of value only if the animal is zinc-deficient. There is no evidence that supplementation of zinc for zinc-adequate animals improves wound healing.

ZINC IN METALLOPROTEINS

Since 1940, when Keilin and Mann discovered the first zinc metalloenzyme, carbonic anhydrase, the list of enzymes in which the functional or structural role of Zn(II) has been documented has increased considerably. Data on approximately 50 enzymes suggest that zinc may be involved as a necessary cofactor. Several metabolically important reactions are catalyzed by these enzymes, including hydrolysis, hydration, oxidation-reduction, and group transfer reactions. Detailed physicochemical data on several of the enzymes catalyzing hydrolysis or hydration reactions, e.g., carboxypeptidase A and carbonic anhydrase, show the zinc to function in its capacity as a Lewis acid by withdrawing electrons from a group of the substrate directly coordinated to the metal ion at a site initially occupied by solvent water. Zinc may also generate active, coordinated hydroxide ions at enzyme-active sites. In each case, the function of the Zn(II) as a Lewis acid is clearly only part of a concerted mechanism involving reactive side chains of the proteins in addition to the metal ion. Whereas such interaction may be the role of Zn(II) in the catalytic mechanism of a number of enzymes, the Zn(II) in superoxide dismutase and aspartate transcarbamylase does not appear to interact directly with the substrate. Thus zinc may also function by maintaining the required conformation of a protein or by participating in the binding of effector molecules to allosteric enzymes. If, in addition to its functional role in well-characterized zinc metalloenzymes, zinc is an absolute requirement

for the function of the nucleotidyl transferase enzymes basic to DNA replication and transcription, then the metal's fundamental role in molecular biology and at least some of the molecular reasons for the severe physiologic effects of zinc deficiency become clear.

CLINICAL ASPECTS OF ZINC METABOLISM

Because zinc is a cofactor in protein synthesis, it plays an active role in many disease processes.

Severe liver disease is commonly associated with loss of total body zinc. In animals, zinc pretreatment has been associated with some protection from liver damage with several hepatotoxins. However, the role of zinc therapy in human liver disease is unclear. Gastrointestinal malabsorption from any cause is associated with decreased gut absorption of zinc and with the subsequent production of zinc deficiency. Although rare, acrodermatitis enteropathica is a severe systemic disease in which zinc malabsorption plays a role in pathogenesis. The symptoms of this disease disappear following exogenous zinc administration.

Serum concentrations of zinc vary with several disease states, including several cancers, blood dyscrasias, infectious processes, and renal diseases. Various drugs alter zinc levels in blood and urine, as do peritoneal and blood dialyses of patients with uremia. Parenteral hyperalimentation is associated with decreased serum zinc concentration and increased urinary zinc excretion, a condition consistent with the production of zinc depletion.

Zinc deficiency, as evidenced by total body loss of zinc, has been produced acutely in man following the oral administration of L-histidine; the symptoms produced by this body depletion of zinc are quickly obviated following exogenous oral zinc administration, even in the face of continued histidine administration.

In animals, several studies have suggested that zinc may inhibit the growth of specialized tumors. There are no applicable data for humans, but changes in serum zinc concentrations accompany many malignant processes at various stages,

Preliminary reports of the usefulness of zinc in the treatment of various medical disorders have appeared but have yet to be substantiated by controlled clinical trials. Claims have been made that schizophrenia, atherosclerotic cardiovascular disease, laryngeal granulomas, gastric ulcers, and some forms of color blindness can be improved by oral administration of zinc ion.

Zinc has been used as a therapeutic preparation for several centuries. It has been applied topically to treat skin disorders since ancient times and is still used for this purpose. Its capacity to produce gastrointestinal irritation is consistent with its use as an emetic. Oral administration of zinc has been used to assist in the more rapid healing of wounds of various types, but this therapy appears to be useful only in patients with signs and symptoms of zinc deficiency.

Zinc has also been used to treat patients with taste and smell dysfunction of various types and, as in the wound healing studies, it is apparently of little value except in those patients with zinc deficiency. Because zinc deficiency may be difficult to ascertain reliably, identification of patients at risk poses a difficult practical problem.

TOXICITY OF ZINC

Humans

Zinc is not a highly toxic substance. Zinc toxicosis may occur only when very high dose levels overwhelm the homeostatic mechanisms controlling zinc uptake and excretion. Reports of zinc tolerance as well as toxicosis in humans are sparse, but they do suggest that 500 mg to 1 g or more may be ingested on a daily basis without adverse effects. Ten or more g taken as a single oral dose may produce gastrointestinal distress, including nausea, vomiting, and diarrhea. There are also suggestions in the literature that even higher dosage may produce dizziness and perhaps increase blood levels of pancreatic enzymes.

None of these observations of acute disorders have been rigorously studied under controlled conditions nor are they confirmed experimentally. Chronic zinc toxicosis in humans is even less well documented.

Inhalation of zinc has been related to metal fume fever, an acute disability of short duration that can occur when fume is inhaled from metal heated to a temperature above its melting point. It is most commonly associated with inhalation of zinc oxide fume and is most severe among brass founders. It is characteristized by hyperpnea, shivering accompanied by fever, profuse sweating, pain in chest and legs, and general weakness beginning 4-8 h after exposure and lasting about 24-48 h. With repeated exposures, some degree of tolerance may be built up, but it will be lost when exposure to fume ceases for a period as short as two days. The pathogenesis of this disorder, including the role of zinc in it, is not understood.

Animals

Although animals have a high tolerance for zinc, problems have been reported in animals (particularly horses) grazing near lead-zinc smelters. Zinc intake of the order of 3,000 ppm (mg/kg body weight) was required to induce the symptoms experimentally. Most animals appear to tolerate levels up to 1,000 ppm in the diet without ill effects if the diet contains adequate copper and iron. High levels of zinc interfere with metabolism of these minerals; therefore anemia and increased serum cholesterol are likely if copper or iron intake is low.

Interactions Between Zinc and Cadmium

Zinc and cadmium have some physiochemical properties in common. There are, however, large differences between these two metals in biologic systems: zinc is essential and has a short biologic half-time, and cadmium is not essential and has an extremely long biologic half-time. Zinc is found in high concentrations in most tissues, whereas cadmium mainly accumulates in kidney and liver. The cadmium- and zinc-binding protein, metallothionein, is the main storage protein for cadmium. Zinc easily crosses the placental barrier, whereas cadmium is practically excluded. Experiments with animals, in which cadmium concentrations in the diet often have been similar to or higher than the zinc concentrations, have shown that cadmium may accentuate symptoms brought on by zinc deficiency. Whereas zinc can counteract some actions of cadmium (such as weight loss), it does not influence anemias caused by cadmium.

Exposure to cadmium will increase zinc levels in kidney and liver. When zinc intake is marginal, exposure to cadmium may cause some tissues, such as testes, to become depleted of zinc. Cadmium has been shown to

interfere with many enzymes both *in vivo* and *in vitro*. Zinc may prevent some of these interferences, but not all.

Limited data exist on zinc-cadmium interactions in human beings. The normal accumulation of cadmium in renal cortex will be accompanied by an equimolar increase in zinc, probably reflecting the metal content of metallothionein.

From the available data, it can be concluded that for human beings who ingest enough zinc and who are not excessively exposed to cadmium, the main metal-metal interaction will take place in the kidneys, where about one-third of the total body burden of cadmium is stored. The cadmium concentrations in other parts of the body are probably too low to interfere greatly with zinc-dependent systems. As long as the amount of zinc necessary for normal function is not altered in the kidneys, cadmium should not cause any functional disturbances in that organ. Higher exposure to cadmium--from industry or food--will elevate cadmium levels and eventually cause renal damage. The few available data indicate that zinc stops increasing when cadmium levels become very high in the renal cortex.

If zinc intake is marginal, then it can be postulated that zinc levels of some tissues may become depressed if the accumulation of cadmium in kidney and liver results in zinc storage there. A risk may be incurred for the fetus if pregnant women are exposed to cadmium without adequate zinc intakes. The danger lies not in a direct action of cadmium, but in the smaller amount of zinc available for the fetus.

Usually animal products rich in protein are a good source for zinc, and cadmium concentrations are very low in meat. If vegetable protein sources replace meat, it can be expected that the zinc:cadmium ratios will not be so favorable.

The World Health Organization has recommended that the weekly intake of cadmium in adults should not exceed 400-500 μg. This means a daily limit of about 70 μg of cadmium. It has also recommended that the concentration in drinking water should not exceed 5 μg cadmium/kg. If zinc supplements are added to human food, care must be taken that excessive amounts of cadmium are not added as well. Since not more than 10 μg (assuming 2 kg water/day) should be allowed from drinking water, less should be allowed from supplements. If zinc is added to human diets, supplements should not cause an increase of more than 5 μg cadmium/day. There is thus need for strict control of zinc compounds used as food additives.

STANDARDS FOR ZINC LEVELS

Standards for zinc in air and water have been obtained using techniques which do not fully reflect available advanced technology. Zinc is ubiquitous and, as such, will be present in the environment forever. Continuous monitoring of the environment is important to maintain a close check on the levels of zinc in air, water, and land.

SAMPLING AND MEASUREMENT TECHNIQUES FOR ANALYZING ZINC

Analysis of Samples from the General Environment

Methods for evaluating zinc in media such as physiologic fluids, soft tissue, bone, hair, waters, soils, foodstuffs, and plants vary greatly in nature as to merits and disadvantages. Techniques include absorption spectrophotometry, atomic absorption spectrophotometry, neutron activation analysis, anodic stripping voltammetry, X-ray fluorescence, arc-emission spectrography, spectrofluorometry, and optical spectrography. Comparatively reliable and standardized methods for zinc in waters, soils, foodstuffs, and plants exist, but for biologic media, recommended routine methodology exists only for urine and serum.

Methods of expressing zinc levels in various media vary considerably. They include wet weight basis, dry weight basis, dry-ash weight basis, and per-unit biochemical reference such as protein, DNA, etc.

Analysis of Zinc in the Air

Modern methods for determining zinc in the atmosphere include absorption spectrophotometry, atomic absorption spectrometry, optical emission spectrography, X-ray fluorescence spectrometry, spark-source mass spectrometry, instrumental neutron activation analysis, and voltammetry (polarography). The inclusion of activation analysis indicates the ready availability of nuclear reactors for carrying out routine analyses. Ring oven techniques have not been discussed because the results obtained are semiquantitative.

The variety of analytic methods and sampling procedures that have been used complicates comparing results of different investigators necessary to coordinate data to test theories of zinc sources, atmospheric transformations, or removal mechanisms. Because the variations reported are a consequence of differences in sample handling and instrument operating procedures, results are difficult if not impossible to interpret. Comparisons of data from different laboratories could lead to erroneous conclusions.

CHAPTER 15

RECOMMENDATIONS

1. Areas of zinc deficiency and zinc excess in soils and waters in the United States should be identified.

2. A systematic program of monitoring air for zinc should be established.

   The sparseness of data on airborne zinc frustrates attempts to quantify and identify sources of zinc in the environment. A study designed to monitor airborne zinc levels both in the general community and near known point sources would greatly assist in determining the environmental impact of diverse zinc sources.

3. Efforts should be made to control zinc levels in zinc-containing wastewaters to avoid excessive contamination of sludges that will be added to agricultural lands.

   Many industrial and sewage sludges contain high levels of zinc. Since such sludges are sometimes used as fertilizers to provide nitrogen, phosphorus, and micronutrients, efforts should be made to ensure that the addition of zinc to soil is not great enough to cause toxicoses in plants and excessive levels in feed and food. Contaminated sludges should be disposed of in sanitary landfills or incinerated; they should not be applied to land indiscriminately.

4. Additional research should be conducted on the occurrence and characteristics of organic zinc complexes in natural waters.

   Chemical complexing of zinc may affect the biologic availability of the metal to aquatic organisms. Naturally occurring physicochemical forms of zinc should be distinguished in order to understand the mechanisms controlling zinc accumulation and metabolism in aquatic organisms.

5. Research is needed on the relation of zinc to aquatic plants.

   a. A broader range of zinc values and concentration factors are needed for many aquatic plant species to evaluate the impact of increasing doses and varied forms of zinc in ambient waters. Sampling and analysis techniques need to be refined to establish critical or threshold values for zinc toxicity and the overall zinc nutrition status in aquatic plants.

   b. The influence of biologic, chemical, and environmental factors in sediments and waters upon zinc content and movement into ambient waters, aquatic plants and ultimately the food chain should be studied further.

   c. The accumulated wealth of information on the functions of zinc in aquatic plants, especially green algae, should be used as a basis for more concentrated research on zinc in plant metabolism. Such an approach would be the most useful for expanding understanding of functions of zinc in plants.

6. Research is needed on the relation of zinc to terrestrial plants.

   a. The specific mechanisms of zinc uptake and translocation at the molecular level need to be identified.

b. Additional metabolic functions need to be characterized, because the roles identified to date require only a small fraction of the zinc needed for normal plant growth.

c. The physiologic basis for differential zinc requirements among species needs to be defined.

d. The mechanism of zinc-phosphorus interactions in plants needs to be determined.

e. The chemical forms of zinc in soils controlling zinc levels in soil solution need to be determined. If this is done, soil solution levels can be monitored for normal plant growth and nutrition and deficiency and toxicosis can be more easily guarded against.

f. Specific mechanisms of zinc deficiencies, toxicoses and tolerance in plants need to be identified.

g. The processes of zinc redistribution within plants, especially the movement from vegetative tissue to seeds, should be determined.

h. The basis of foliar absorption and movement of zinc in plants needs to be established.

7. Experiments on acute and chronic exposure of aquatic organisms to elevated zinc concentrations in aqueous systems should be undertaken on a systematic basis to determine the effects of zinc on vital life processes. The effects of zinc should be determined alone and in combination with other potential contaminants of human origin, such as cadmium. Studies should be conducted for various critical life stages under many combinations of important environmental interactions (salinity, temperature, pH, dissolved oxygen) over all ranges of

variation to which the test organism is exposed naturally. Such a systematic approach is required to ensure adequate water quality standards for protecting aquatic organisms.

8. Research into the physiologic role of zinc in humans should be encouraged. Systems to be investigated should include the endocrine system, sensory systems, including vision and taste, muscle and the nervous system, including the central and peripheral nervous systems and behavior.

9. The following additional research on zinc in the diet is needed:

   a. A method of assessing zinc status of man and animals. There is no currently accepted method of determining whether or not an animal is receiving adequate zinc.

   b. The occurrence of marginal zinc deficiencies. Because of the lack of a method of determining zinc status, the prevalence of marginal zinc deficiency is completely unknown.

   c. The requirement for zinc and factors that affect the requirement. The data available for determining the amount of zinc required, particularly by man, are very limited. Since diets low in protein will not generally contain the 15 mg zinc recommended daily for adult humans, it is important to know how low an intake would be adequate. It has been suggested that additional zinc may be beneficial for both man and animals in times of stress, but more information is needed.

d. Factors affecting the availability of zinc. Reports of zinc deficiency in animals, particularly ruminants, fed diets containing presumably adequate zinc indicate the need for additional information on factors affecting the metal's availability. Almost no information exists on constituents of plants, other than phytate, that affect the availability of zinc.

e. Content of zinc in food. Attempts to calculate the dietary zinc intake in man are hampered by the lack of adequate or accurate information in the literature. Additional data could be obtained readily with methods now available.

f. Zinc and cadmium. Zinc requirements should be studied in human populations exposed to excessive cadmium. More studies should be conducted on the effect of prolonged exposure to small amounts of cadmium on zinc metabolism of animals on deficient, marginal, and zinc-adequate diets. Zinc metabolism and its relation to cadmium exposure in pregnancy especially should be studied.

10. <u>The role, if any, of zinc in cancer should be investigated systematically</u>. This examination should include an evaluation of changes in zinc metabolism that occur in various cancers and the role, if any, of zinc as an inhibitor of cancer growth.

11. <u>The role, if any, of zinc deficiency in human fetal wastage and congenital malformations should be investigated</u>.

12. <u>Efficacy of zinc therapy for various diseases should be evaluated through controlled, randomized, cooperative studies</u>.

Such trials could deal with the question of zinc treatment in wound healing, anorexia, and taste and small dysfunction.

13. Cadmium levels should be evaluated in preparations in zinc used as foodstuffs.

14. Standard reference materials and standard analytic methods for atmospheric particulates should be developed.

Such measures would permit analysts to check their results. The availability of standard materials and procedures would provide a means not only to evaluate the reliability of analytic methods, but also to compare data obtained in different laboratories from different samples at different times.

APPENDIX A

Zinc Content of Foods[a]

I.

Food and Description[b]	Zinc, mg[c]
Apples, raw	0.05
Applesauce, unsweetened	0.1
Bananas, raw	0.2
Beans, common, mature, dry	
Raw	2.8
Boiled, drained	1.0
Beans, lima, mature, dry	
Raw	2.8
Boiled, drained	0.9
Beans, snap, green	
Raw	0.4
Boiled, drained	0.3
Canned, solids and liquid	0.2
Canned, drained solids	0.3
Beef, separable lean	
Raw	4.2
Cooked, dry heat	5.8
Cooked, moist heat	6.2
Beef, separable fat, raw	0.5
Beef, ground (77% lean)	
Raw	3.4
Cooked	4.4
Beverages, carbonated, nonalcoholic	
Bottled	<0.01
Canned	0.08
Bran, see wheat	
Breads	
Rye	1.6
White	.6
Whole wheat	1.8
Butter	0.1
Cabbage, common	
Raw	0.4
Boiled, drained	0.4
Cake, white, without icing	0.2
Carrots	
Raw	0.4
Cooked or canned, drained solids	0.3
Cheese, cheddar type	4.0
Chicken, broiler-fryer	
Breast, meat only	
Raw	0.7
Cooked, dry heat	0.9
Breast	
Raw (81% meat, 12% skin, 7% fat)	0.7
Cooked, dry heat (89% meat,11% skin)	0.9
Drumstick, thigh, back, meat only	
Raw	1.8
Cooked, dry heat	2.8
Drumstick	
Raw (85% meat, 13% skin, 2% fat)	1.7
Cooked, dry heat (84% meat,16% skin)	2.5
Wing, meat only	
Raw	1.6
Cooked, dry heat	2.4
Neck, meat only	
Raw	2.7
Cooked, moist heat	3.0
Skin	
Raw	1.0
Cooked, dry heat	1.2

APPENDIX A - continued

I.

Food and Description[b]	Zinc, mg[c]
Chickpeas or garbanzos, mature seeds, dry	
Raw	2.7
Boiled, drained	1.4
Chocolate syrup	0.9
Clams	
Soft shell	
Raw	1.5
Cooked	1.7
Hard shell	
Raw	1.5
Cooked	1.7
Surf, canned, solids and liquid	1.2
Cocoa, dry powder	5.6
Coffee	
Dry, instant	0.6
Fluid beverage	0.03
Cookies, vanilla wafers	0.3
Cooking oil, see oils	
Corn, field, whole-grain, yellow, or white	2.1
Corn, sweet, yellow	
Raw	0.5
Boiled, drained	0.4
Corn, canned, whole kernel, yellow	
Brine pack, solids and liquid	0.3
Brine pack, drained solids	0.4
Vacuum pack, solids and liquid	0.4
Corn chips	1.5
Corn grits, white, degermed, dry form	0.4
Corn flakes	0.3
Cornmeal, white or yellow	
Bolted (nearly whole grain)	1.8
Degermed	
Dry form	0.8
Cooked	0.1
Cornstarch	0.03
Cowpeas (blackeyed), mature, dry	
Raw	2.9
Boiled, drained	1.2
Crabs, blue and Dungeness	
Raw	4.0
Steamed	4.3
Crackers	
Graham	1.1
Saltines	0.5
Doughnuts, cake-type	0.5
Eggs, fresh	
Whites	0.02
Yolks	3.0
Whole	1.0
Farina, regular	
Dry form	0.5
Cooked	0.06
Fish, white varieties, flesh only	
Raw	0.7
Cooked, fillet	1.0
Cooked, steak	0.8
Gizzard	
Chicken	
Raw	2.9
Cooked, drained	4.3

APPENDIX A - continued

I.

Food and Description[b]	Zinc, mg[c]
Gizzard	
Turkey	
Raw	2.8
Cooked, drained	4.1
Granola	2.1
Heart	
Chicken	
Raw	2.9
Cooked, drained	4.8
Turkey	
Raw	2.8
Cooked, drained	4.8
Ice Cream	0.5
Lamb	
Separable lean	
Raw	3.0
Cooked, dry heat	4.3
Cooked, moist heat	5.0
Separable fat, raw	0.5
Lard	0.2
Lentils, mature, dry	
Raw	3.1
Boiled, drained	1.0
Lettuce, head or leaf	0.4
Liver	
Beef	
Raw	3.8
Cooked	5.1
Calf	
Raw	3.8
Cooked	6.1
Liver	
Chicken	
Raw	2.4
Cooked	3.4
Turkey	
Raw	2.7
Cooked	3.4
Lobster, crayfish	
Raw	1.8
Cooked or canned	2.2
Macaroni	
Dry form	1.5
Cooked, tender stage	0.5
Margarine	0.2
Milk	
Fluid, whole or skim	0.4
Canned, evaporated	0.8
Dry, nonfat	4.5
Oatmeal or rolled oats	
Dry form	3.4
Cooked	0.5
Oat cereal, puffed, ready-to-eat	3.0
Oil, salad or cooking	0.2
Onions, mature or green, raw	0.3
Oranges, raw	0.2
Orange juice	
Canned, unsweetened	0.07
Fresh or frozen	0.02
Oysters, raw or frozen	
Atlantic	74.7
Pacific	9.0

APPENDIX A - continued

I.

Food and Description[b]	Zinc, mg[c]
Peaches	
Raw	0.2
Canned, drained slices	0.1
Peanuts	
Raw	2.9
Roasted	3.0
Peanut butter	2.9
Peas, green, immature	
Raw	0.9
Boiled, drained	0.7
Canned, drained solids	0.8
Peas, green, mature seeds, dry	
Raw	3.2
Boiled, drained	1.1
Popcorn	
Unpopped	3.9
Popped	
Plain	4.1
Oil and salt added	3.0
Pork	
Trimmed lean cuts, separable lean	
Raw	2.7
Cooked	3.8
Boston butt, separable lean	
Raw	3.2
Cooked	4.5
Ham or picnic, separable lean	
Raw	2.8
Cooked	4.0
Pork	
Loin, separable lean	
Raw	2.2
Cooked	3.1
Separable fat, raw	0.5
Potatoes	
Raw	0.3
Boiled, drained	0.3
Rice	
Brown	
Dry form	1.8
Cooked	0.6
White, regular	
Dry form	1.3
Cooked	0.4
White, parboiled	
Dry form	1.1
Cooked	0.3
White, precooked, quick	
Dry form	0.7
Cooked	0.2
Cereal, ready-to-eat, puffed or flakes	1.4
Rolls, hamburger	0.6
Salad dressing	0.2
Salmon, canned (77% solids, 23% liquid)	0.9
Sausages and cold cuts	
Bologna, beef	1.8
Braunschweiger	2.8
Frankfurters	
Made with beef	2.0
Made with beef and pork	1.6

I.

Food and Description[b]	Zinc, mg[c]
Shrimp	
Raw	1.5
Boiled, peeled, deveined	2.1
Canned, drained solids	2.1
Spinach	
Raw	0.8
Boiled, drained	0.7
Canned	
Solids and liquid	0.6
Drained solids	0.8
Sugar, white, granulated	0.06
Tea	
Dry leaves	3.3
Fluid beverage	0.02
Tomatoes, ripe	
Raw	0.2
Boiled, solids and liquid	0.2
Canned, solids and liquid	0.2
Tunafish, canned in oil (85% solids, 15% oil)	1.0
Drained solids	1.1
Turkey	
Light meat	
Raw	1.6
Cooked, dry heat	2.1
Dark meat	
Raw	3.1
Cooked, dry heat	4.4
Neck meat	
Raw	5.0
Cooked	6.4

Food and Description[b]	Zinc, mg[c]
Turkey	
Skin	
Raw	1.3
Cooked	2.1
Veal	
Separable lean	
Raw	2.8
Cooked, dry heat	4.1
Cooked, moist heat	4.2
Separable fat, raw	0.5
Wheat, whole grain	
Hard	3.4
Soft	2.7
White	2.2
Durum	2.7
Wheat flours	
Whole	2.4
80% extraction	1.5
All-purpose	0.7
Bread flour	0.8
Cake or pastry flour	0.3
Wheat bran, crude	9.8
Wheat germ, crude	14.3
Wheat cereal, whole-meal	
Dry form	3.6
Cooked	0.5

APPENDIX A - continued

I.

Food and Description[b]	Zinc, mg[c]
Wheat cereals, ready-to-eat	
Bran flakes, 40%	3.6
Flakes	2.3
Germ, toasted	15.4
Puffed	2.6
Shredded	2.8

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[a]Reproduced with permission from Murphy et al.

[b]100 g, edible portion.

[c]Data are given to two decimal places if food contains less than 0.1 mg zinc per edible portion.

APPENDIX A

Zinc Content of Foods[a]

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Apples, raw	1 medium	180	0.08
Applesauce, unsweetened	1 cup	244	0.3
Bananas, raw	1 medium	119	0.3
Beans, common, mature dry			
Raw	1 cup	190	5.3
Boiled, drained	1 cup	185	1.8
Beans, lima, mature, dry			
Raw	1 cup	180	5.0
Boiled, drained	1 cup	190	1.7
Beans, snap, green			
Raw, cut into 1-2 in. lengths	1 cup	110	0.4
Boiled, drained, cut and French style	1 cup	125	0.4
Canned, solids and liquid	1 cup	239	0.6
Canned, drained solids	1 cup	135	0.4
Beef, separabe lean			
Cooked, dry heat	3 oz	85	4.9
Cooked, moist heat	3 oz	85	5.3
Beef, ground, cooked	3 oz	85	3.8
Beverages, carbonated, non-alcoholic			
12 fl oz (360 ml)	1 bottle	367	0.01
12 fl oz	1 can	367	0.3
Breads			
Rye	1 slice	25	0.4
White	1 slice	28	0.2
Whole wheat	1 slice	28	0.5
Butter, 4 sticks/lb	1 cup	227	0.2
	1 tbsp	14	0.01
Cabbage, common			
Raw, shredded finely	1 cup	90	0.3
Shredded, boiled, drained	1 cup	145	0.6
Cake, white, without icing (3 x 3 x 2 in.)	1 piece	86	0.2
Carrots			
Raw	1 medium	72	0.3
Cooked or canned, drained solids	1 cup	155	0.5
Cheese, cheddar	1 slice	13	0.5
Chicken, broiler-fryer, cooked, dry heat			
Breast, cooked			
Meat only	1/2 breast	85	0.7
Meat and skin	1/2 breast	96	0.9

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Chicken, broiler-fryer, cooked, dry heat			
Drumstick, thigh, back, meat only, cooked	3 oz	85	2.4
Drumstick			
Meat only	1 drumstick	45	1.3
Meat and skin	1 drumstick	54	1.4
Chickpeas, mature, dry			
Raw	1 cup	200	5.4
Boiled, drained	1 cup	146	2.0
Chocolate syrup, 1 fl oz	2 tbsp	38	0.3
Clams			
Soft shell, cooked	3 oz	85	1.4
Hard shell			
Raw	4 cherrystones or 5 littlenecks	70	1.1
Cooked	4 or 5 clams	62	1.0
Surf, canned, solids and liquids, can size 211 x 300	1 can	220	2.7
Cocoa, dry powder			
Approx 5 1/4 tbsp	1 oz	28	1.6
Coffee			
Dry, instant	1 tbsp	2.5	0.02
Fluid beverage, 6 fl oz	1 cup	180	0.05
Cookies (1 3/8 x 1/4 in)	10 cookies	30	0.08
Corn, sweet, yellow			
Boiled, drained	1 cup	165	0.7
Canned, vacuum pack	1 cup	210	0.8
Corn chips	1 oz	28	0.4
Corn grits, dry form	1 cup	160	0.7
Corn flakes	1 oz	28	0.08
Cornmeal, white or yellow			
Bolted, dry form	1 cup	122	2.1
Degermed			
Dry form	1 cup	138	1.2
Cooked	1 cup	240	0.3
Cowpeas (blackeyes)			
Raw	1 cup	170	4.9
Boiled, drained	1 cup	250	3.0
Crabs, steamed, pieces	1 cup	155	6.7
Crackers			
Graham (2 1/2 x 2 1/2 in)	2 squares	14	0.2
Saltines	10 crackers	28	0.1
Doughnuts (3 1/4 in. diam)	1 doughnut	42	0.2

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Eggs, fresh			
White	1 large	33	<0.01
Yolk	1 large	17	0.5
Whole	1 large	50	0.5
Farina, regular			
Dry form	1 cup	180	1.0
Cooked	1 cup	245	0.2
Fish, white varieties, fresh only			
Fillet, cooked	3 oz	85	0.9
Steak, cooked	3 oz	85	0.7
Gizzard, cooked, drained, diced			
Chicken	1 cup	145	6.2
Turkey	1 cup	145	6.0
Granola	1 oz	28	0.6
Heart, cooked, drained, diced			
Chicken	1 cup	145	6.9
Turkey	1 cup	145	7.0
Ice Cream	1 cup	133	0.6
Lamb, separable lean			
Cooked, dry heat	3 oz	85	3.7
Cooked, moist heat	3 oz	85	4.2
Lard	1 cup	205	0.4
	1 tbsp	13	0.03
Lentils, mature, dry			
Raw	1 cup	190	5.9
Boiled, drained	1 cup	200	2.0
Lettuce, head or leaf			
Approx 1/6 head	1 wedge	90	0.4
Loose leaf, chopped	1 cup	55	0.2
Liver, cooked			
Beef	2 oz	57	2.9
Calf	2 oz	57	3.5
Chicken, chopped	1 cup	140	4.7
Turkey, chopped	1 cup	140	4.7
Lobster, cooked, cubed	1 cup	145	3.1
Macaroni, cooked, tender			
Measured hot	1 cup	140	0.7
Measured cold	1 cup	105	0.5
Margarine	1 cup	227	0.5
	1 tbsp	14	0.03
Milk			
Fluid	1 cup	244	0.9
Canned, evaporated	1 cup	252	1.9
Dry, nonfat	1 cup	68	3.1

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Oatmeal or rolled oats			
Dry form	1 cup	80	2.7
	1 oz	28	1.0
Cooked	1 cup	240	1.2
Oat cereal, puffed	1 oz	28	0.8
Oil, salad or cooking	1 cup	218	0.4
Onions			
Mature, chopped	1 cup	170	0.6
Young green, chopped	1 cup	100	0.3
Oranges, raw, 2 5/8 in. diam	1 orange	131	0.2
Orange juice			
Canned, unsweetened	1 cup	249	0.2
Fresh or frozen	1 cup	248	0.05
Oysters			
Atlantic			
Raw, drained, 12 fl oz can, 18-27 select or 27-44 standard oysters	1 can	340	254.3
Frozen, solids and liquid, 12 fl oz can	1 can	360	268.9
Pacific			
Raw, drained, 12 fl oz can, 6-9 medium or 9-13 small oyster	1 can	340	30.6
Frozen, solids and liquid, 12 fl oz can	1 can	360	32.4
Peaches			
Raw, peeled, 2 1/2 in. diam	1 medium	100	0.2
Canned, drained, slices	1 cup	220	0.3
Peanuts, roasted	1 tbsp	9	0.3
Peanut butter	1 tbsp	16	0.5
Peas, green, immature			
Raw or frozen	1 cup	145	1.2
Boiled, drained	1 cup	160	1.2
Canned, drained solids	1 cup	170	1.3
Peas, green, mature seeds, dry			
Raw	1 cup	200	6.4
Boiled, drained	1 cup	200	2.1
Popcorn			
Unpopped	1 cup	205	7.9
Popped			
Plain, large kernel	1 cup	6	0.2
With oil and salt	1 cup	9	0.3

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Pork, cooked, dry heat, separable lean			
Trimmed lean cuts	3 oz	85	3.2
Boston butt	3 oz	85	3.8
Ham or picnic	3 oz	85	3.4
Loin	3 oz	85	2.6
Potatoes			
Raw, peeled, 2 1/2 in. diam	1 medium	112	0.4
Pared before cooking, boiled, drained	1 medium	112	0.3
Boiled in skin, drained, pared	1 medium	136	0.4
Rice			
Brown			
Dry form	1 cup	185	3.4
Cooked, measured hot	1 cup	195	1.2
White, regular, long-grain			
Dry form	1 cup	185	2.5
Cooked			
Measured hot	1 cup	205	0.8
Measured cold	1 cup	145	0.6
White, parboiled			
Dry form	1 cup	185	2.1
Cooked			
Measured hot	1 cup	175	0.6
Measured cold	1 cup	145	0.5
White, precooked, quick			
Dry form	1 cup	95	0.7
Cooked			
Measured hot	1 cup	165	0.4
Measured cold	1 cup	130	0.3
Cereal, ready-to-eat, puffed/flaked	1 oz	28	0.4
Rolls, hamburger, 3 1/2 in. diam	1 roll	40	0.2
Salad dressing	1 tbsp	15	0.03
Salmon, canned, solids and liquid	1 cup	220	2.1
Sausages and cold cuts			
Bologna, beef, 4 1/2 in. diam, 1 oz	1 slice	28	0.5
Braunschweiger, 1 oz	1 slice	28	0.8
Frankfurters			
Made with beef, 10 per lb	1 frank	45	0.9
Made with beef and pork, 10 per lb	1 frank	45	0.7

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Shrimp			
Boiled, peeled, deveined, 33 per lb	6 shrimp	84	1.7
Canned, drained, solids	1 cup	128	2.7
Spinach			
Raw, chopped	1 cup	55	0.5
Boiled, drained	1 cup	180	1.3
Canned			
Solids and liquid	1 cup	232	1.5
Drained solids	1 cup	205	1.6
Sugar, white, granulated	1 cup	200	0.1
Tea, fluid beverage, 6 fl oz	1 cup	177	0.04
Tomatoes, ripe			
Raw, 2.6 in.diam	1 medium	123	0.2
Boiled	1 cup	241	0.5
Canned, solids and liquid	1 cup	241	0.5
Tunafish, canned in oil			
Chunk style, solids and liquid, can size 307 x 113, 6 1/2 oz	1 can	184	1.7
Drained solids, can size 307 x 113, 6 1/2 oz	1 can	157	1.8
Drained solids	1 cup	160	1.8
Turkey, cooked, dry heat, meat only			
Light meat	3 oz	85	1.8
Dark meat	3 oz	85	3.7
Veal, separable lean			
Cooked, dry heat	3 oz	85	3.5
Cooked, moist heat	3 oz	85	3.6
Wheat flours			
Whole, stirred, spooned into cup	1 cup	120	2.9
All purpose, sifted, spooned into cup, standard granulation	1 cup	115	0.8
Bread flour, sifted, spooned into cup, standard granulation	1 cup	115	0.9
Cake flour, sifted, spooned into cup	1 cup	96	0.3
Wheat cereal, wholemeal			
Dry form	1 cup	125	4.5
Dry form	1 oz	28	1.0
Cooked	1 cup	245	1.2
Cooked, from 1 oz dry		216	1.0

II.

Food	Approximate Measure	Weight, g[b]	Zinc, mg[c]
Wheat cereals, ready-to-eat			
Bran flakes, 40%	1 oz	28	1.0
Flakes	1 oz	28	0.6
Germ, toasted	1 tbsp	6	0.9
Puffed	1 oz	28	0.7
Shredded	1 oz	28	0.8

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[a] Reproduced with permission of Murphy et al.

[b] Edible part of common household units; measure and weight only apply to edible part of food.

[c] Data are given to two decimal places if food contains less than 0.1 mg zinc per edible portion.

APPENDIX B

Methods of Zinc Analysis*

ZINC

Zinc is an essential and beneficial element in body growth. However, concentrations above 5 mg/l can cause a bitter astringent taste and an opalescence in alkaline waters. The zinc concentration of U.S. drinking waters varies between 0.06 and 7.0 mg/l, with a mean of 1.33 mg/l. Zinc most commonly enters the domestic water supply from the deterioration of galvanized iron and the dezincification of brass. In such cases the presence of lead and cadmium also may be suspected, because they are impurities of the zinc used in galvanizing. Zinc also may result from industrial waste pollution.

1. Selection of Method

Where the equipment is available, the atomic absorption spectrophotometric method is preferred for the determination of zinc. The dithizone and zincon colorimetric methods are useful in the absence of the sophisticated instrumentation. Dithizone method I is intended for unpolluted waters, and II for polluted waters or wastewater.

2. Sampling and Storage

Analyze samples within 6 hr after collection. The addition of HCl will preserve the metallic ion content but requires that: (*a*) the acid be zinc-free; (*b*) the sample bottles be rinsed with acid before use; and (*c*) the samples be evaporated to dryness in silica dishes to remove the excess HCl before analysis.

Atomic Absorption Spectrophotometric Method

1. General Discussion

a. Principle: Atomic absorption spectrophotometry resembles emission flame photometry in that a sample is aspirated into a flame and atomized. The major difference is that flame photometry measures the amount of light emitted, whereas in atomic absorption spectrophotometry a light beam is directed through the flame, into a monochromator, and onto a detector that measures the amount of light absorbed by the atomized element in the flame. For many metals difficult to analyze by flame emission, atomic absorption exhibits superior sensitivity. Because each metal has its own characteristic absorption wavelength, a source lamp composed of that element is used; this makes the method relatively free from spectral or radiation interferences. Thus the amount of energy of the characteristic wavelength absorbed in the flame is proportional to the concentration of the element in the sample.

b. Interference: Most metals can be determined by direct aspiration of the sample into an air-acetylene flame. The most troublesome type of interference is termed "chemical" and results from the lack of absorption of atoms bound in molecular combination in the flame. This can occur when the flame is not hot enough to dissociate the molecules (in the case of phosphate interference with magnesium) or when the dissociated atom is oxidized immediately to a compound that will not dissociate further at the temperature of the flame. The interference of phosphate in the magnesium determination can be overcome by the addition of lanthanum. Similarly, the introduction of calcium eliminates silica interference in the determination of manganese. However, silicon and metals such as aluminum, barium, beryllium, and vanadium require the use of the higher-temperature, nitrous oxide-acetylene flame to dissociate their molecules. In addition, barium undergoes ionization in the flame and the ground state (potentially absorbing) population is thereby reduced. The addition of an excess of a cation (sodium or potassium) having a similar or lower ionization potential will overcome this problem. The wavelength of maximum absorption for arsenic is 193.7 nm, and for selenium 196.0 nm. Unfortunately, the air-acetylene flame absorbs intensely at these wavelengths. The sensitivity of the method for these metals can be improved by the use of the argon-hydrogen flame. In the determination of mercury by the cold vapor (flameless) technic, certain volatile organic materials may absorb at 253.7 nm. If this is expected, the sample should be analyzed by the regular procedure and again under oxidizing conditions only, that is, without the addition of stannous chloride. The true mercury concentration can be obtained by subtracting the two values.

c. Sample handling: Before collecting a sample, decide on the type of data desired, i.e., dissolved, suspended, total, or extractable metals. This decision will determine whether the sample is to be acidified, with or without filtration, and the kind of digestion required.

Acidify all samples at the time of collection to keep the metals in solution and to minimize their adsorption on the container wall. If only dissolved metals are to be measured, filter the sample through a 0.45-μm membrane before acidification. If possible, filter and acidify in the field at the time of collection. Report the results obtained on this sample as "dissolved." Filtration is not necessary when total or extractable concentrations are required.

Acidify the sample with conc HNO_3 to a pH of 2.0 or less. Usually, 1.5 ml conc HNO_3/l sample will be sufficient for potable waters free from particulate matter. Such samples can be analyzed with no further treatment. However, samples containing suspended materials or organic matter require pretreatment, as described below.

2. Apparatus

a. Atomic absorption spectrophotometer, consisting of a source of light emitting the line spectrum of an element (hollow cathode lamp), a device for vaporizing the sample (usually a flame), a means of isolating an absorption line (monochromator or filter and adjustable slit), and a photoelectric detector with its associated amplifying and electronic measuring equipment. Both direct current and alternating current systems are used in atomic absorption instruments. The AC or chopped-beam system is preferred because with this system flame emission can be distinguished from lamp emission. For waters high in salt, the use of either a deuterium background corrector or a double-beam instrument that permits the measurement of the absorption at two different wavelengths simultaneously may be helpful.

b. Burner: The most common type of burner is known as a premix, which introduces the spray into a condensing chamber for removal of large droplets. The burner may be fitted with a conventional head containing a single slot 7.6 cm (3 in.) long, which is most useful for aspiration when organic solvents are used; a three-slot Boling head, which is preferred for direct aspiration with an air-acetylene flame; or a head containing a single slot 5 cm (2 in.) long for use with nitrous oxide and acetylene.

c. Recorder: While most instruments are equipped with either a digital or null meter readout mechanism, a good-quality 10-mV recorder with high sensitivity and a fast response time is needed to record the peaks resulting from the determination of mercury by the cold vapor (flameless) technic and for the determination of arsenic and selenium by aspiration of their gaseous hydrides.

d. Hollow cathode lamps: Use one for each element being measured. Multi-element lamps are available but not recommended because they may require the selection of different operating parameters.

e. Pressure-reducing valves: Maintain the supplies of fuel and oxidant at pressures somewhat higher than the controlled operating pressure of the instrument by suitable reducing valves. Use separate reducing valves for each gas.

f. Vent: Place a vent about 15 to 30 cm (6 to 12 in.) above the burner to remove the fumes and vapors from the flame. This precaution protects the laboratory personnel from toxic vapors, prevents the stability of the flame from being affected by room drafts, and protects the instrument from corrosive vapors. A damper or variable-speed blower is also desirable for modulating the air flow and preventing disturbance of the flame.

3. Extractable Metals Analyses

"Extractable metals" include metals in solution plus metals lightly adsorbed on the suspended material. The results obtained in analyses for extractable metals will be influenced by the kind of acid or acids used in the digestion, the concentration of acid, and the heating time. Unless conditions are controlled rigidly, results will be meaningless and unreproducible. The following procedure determines metals soluble in hot HCl-HNO_3. At the time of collection, acidify the entire sample with 5 ml conc HNO_3/l sample. At the time of analysis, mix the sample, transfer a 100-ml portion to a beaker or flask, and add 5 ml 1+1 redistilled HCl. Heat 15 min on a steam bath. Filter and adjust the volume to 100 ml. The sample is then ready for analysis.

The data approximate the total metals in the sample, although something less than the actual total is measured. Concentrations of metal found, especially in heavily silted samples, will be substantially higher than results obtained on only the soluble fraction. Report as "extractable" metals.

Determination of Zinc by Direct Aspiration into an Air-Acetylene Flame

1. Apparatus

See above for a description of the required atomic absorption spectrophotometer and associated equipment. The three-slot Boling burner head is recommended.

2. Reagents

a. Air, cleaned and dried through a suitable filter to remove oil, water, and other foreign substances. The source may be a compressor or commercially bottled gas.

b. Acetylene, standard commercial grade. Acetone, which is always present in acetylene cylinders, can be prevented from entering and damaging the burner head by replacing a cylinder when its pressure has fallen to 7 kg/cm^2 (100 psig) acetylene.

c. Calcium solution: Dissolve 630 mg calcium carbonate, $CaCO_3$, in 10 ml conc HCl. Add 200 ml water, and if necessary heat the solution and boil gently to obtain complete solution. Cool and dilute to 1,000 ml with deionized distilled water.

d. Deionized distilled water: Use deionized distilled water for the preparation of all reagents and calibration standards and as dilution water.

e. Hydrochloric acid, HCl, conc.

f. Lanthanum solution: Dissolve 58.65 g lanthanum oxide, La_2O_3, in 250 ml conc HCl. Add the acid slowly until the material is dissolved and dilute to 1,000 ml with deionized distilled water.

g. Nitric acid, HNO_3, conc.

h. Standard metal solutions: Prepare a series of standard metal solutions containing 5 to 1,000 µg/l by appropriate dilution of the following stock metal solutions with deionized distilled water containing 1.5 ml conc HNO_3/l.

12) *Zinc:* Dissolve 1.000 g zinc metal in 20 ml 1+1 HCl and dilute to 1,000 ml with deionized distilled water; 1.00 ml = 1.00 mg Zn.

3. Procedure

a. Instrument operation: Because of differences between makes and models of satisfactory atomic absorption spectrophotometers, it is not possible to formulate instructions applicable to every instrument. In general, proceed according to the following steps:

1) Install a hollow cathode lamp of the desired metal in the instrument, set the wavelength dial according to Table 301:II, and align the lamp in accordance with the manufacturer's instructions.

2) Set the slit width according to the manufacturer's suggested setting for the element being measured.

3) Turn on the instrument and apply the amount of current suggested by the manufacturer to the hollow cathode lamp.

4) Allow the instrument to warm up until the energy source stablilizes; this process usually requires 10 to 20 min. Readjust the current as necessary after warmup.

5) Install the burner heads.

6) Turn on the air and adjust the flow rate to that specified by the manufacturer to give maximum sensitivity for the metal being measured.

7) Turn on the acetylene, adjust the flow rate to the value specified, and ignite the flame.

8) Atomize deionized distilled water acidified with 1.5 ml conc HNO_3/l, and check the aspiration rate over 1 min. Adjust if necessary to a rate between 3 and 5 ml/min, and zero the instrument.

9) Atomize a standard (usually a 0.5-mg/l standard is suitable) and adjust the burner both up and down and sideways until a maximum response is obtained.

10) The instrument is now ready to operate. When analyses are finished, extinguish the flame by turning off first the acetylene and then the air.

b. Standardization:

1) Select at least three concentrations of each of the standard metal solutions (prepared as in 2*b* above) so as to bracket the expected metal concentration of a sample. Aspirate each in turn into the flame and record the absorbance.

2) For calcium and magnesium calibration, mix 100 ml of standard with 25 ml of lanthanum solution (see 2*f* above) before aspirating.

3) For iron and manganese calibration mix 100 ml of standard with 25 ml of calcium solution (see 2*c* above) before aspirating.

4) With some instruments, it may be necessary to convert percent absorption to absorbance by use of a suitable table generally provided by the manufacturer.

5) Prepare a calibration curve by plotting on linear graph paper the absorbance of the standards versus their concentration.

6) Plot calibration curves for iron and manganese based on the original concentrations of the standards before dilution with calcium solution (¶2*c*).

7) Plot calibration curves for calcium and magnesium based on the original concentration of the standards before dilution with lanthanum solution (¶2*f*).

8) Recheck the calibration curve by aspirating at least one standard after the completion of the analysis of a group of unknown samples.

c. Analysis of samples:

1) Rinse the atomizer by aspirating deionized distilled water containing 1.5 ml conc HNO_3/l, and zero the instrument.

2) Atomize the sample and determine its absorbance.

3) When determining calcium or magnesium, dilute and mix 100 ml sample with 25 ml lanthanum solution (¶2*f*) before atomization.

4) When determining iron or manganese, dilute and mix 100 ml sample with 25 ml calcium solution (¶2*c*) before atomization.

4. Calculations

Calculate the concentration of each metal ion, in micrograms per liter, by referring to the appropriate calibration curve prepared according to 3*b* 5), 6), and 7).

Dithizone Method I

1. General Discussion

a. Principle: Nearly 20 metals are capable of reacting with diphenylthiocarbazone (dithizone) to produce colored coordination compounds. These dithizonates are extractable into organic solvents such as carbon tetrachloride. Most interferences in the zinc-dithizone reaction can be overcome by adjusting the solution to pH 4.0 to 5.5 and by the addition of sufficient sodium thiosulfate. Zinc also forms a weak thiosulfate complex that tends to retard the slow and incomplete reaction between zinc and dithizone. For this reason, the determination is empirical and demands the use of an identical technic in standard and sample analysis. The duration and vigor of shaking, the volumes of sample, sodium thiosulfate, and dithizone, and the pH should all be kept constant.

b. Interference: Interference from bismuth, cadmium, cobalt, copper, gold, lead, mercury, nickel, palladium, silver, and stannous tin in the small quantities found in potable waters is eliminated by complexing with sodium thiosulfate and by pH adjustment. Ferric iron, residual chlorine, and other oxidizing agents convert dithizone to a yellow-brown color. The zinc-dithizone reaction is extremely sensitive, and unusual precautions must be taken to avoid contamination. Experience has shown that high and erratic blanks are often traceable to glass containing zinc oxide, surface-contaminated glassware, rubber products, stopcock greases, reagent-grade chemicals, and distilled water. The extreme sensitivity of the reaction makes it desirable to prepare and segregate glassware especially for this determination and to extract reagents with dithizone solution to remove all traces of zinc and contaminating metals. Dithizone and dithizonates decompose rapidly in strong light. Perform analyses in subdued light and do not expose the solutions to the light of the photometer for a longer period than is necessary. Avoid direct sunlight at all times.

c. Minimum detectable quantity: 1 μg Zn.

2. Apparatus

a. Colorimetric equipment: Use one of the following, although it is also possible to make visual comparisons directly in separatory funnels:

1) *Spectrophotometer,* for use at either 535 or 620 nm, providing a light path of 2 cm.

2) *Filter photometer,* providing a light path of 2 cm and equipped with either a green filter having maximum transmittance near 535 nm or a red filter having maximum transmittance near 620 nm.

3) *Nessler tubes,* matched.

b. Separatory funnels, capacity 125 to 150 ml, Squibb form, preferably with inert teflon stopcocks. If the funnels are of identical size and shape, visual color comparisons may be made directly in them.

c. Glassware: Rinse all glassware with 1+1 HNO_3 and zinc-free water.

d. pH meter.

3. Reagents

a. Zinc-free water: Use redistilled or deionized distilled water for rinsing ap-

paratus and the preparation of solutions and dilutions.

b. Stock zinc solution: Dissolve 100.0 mg 30-mesh zinc metal in a slight excess of 1+1 HCl; about 1 ml is required. Dilute to 1,000 ml with zinc-free water; 1.00 ml=100 μg Zn.

c. Standard zinc solution: Dilute 10.00 ml zinc stock solution to 1,000 ml with zinc-free water; 1.00 ml=1.00 μg Zn.

d. Hydrochloric acid, HCl, 0.02*N:* Dilute 1.0 ml conc HCl to 600 ml with zinc-free water. If high blanks are traced to this reagent, dilute conc HCl with an equal volume of distilled water and redistill in an all-pyrex still.

e. Sodium acetate, 2*N:* Dissolve 68 g $NaC_2H_3O_2 \cdot 3H_2O$ and dilute to 250 ml with zinc-free water.

f. Acetic acid, 1+7. Use zinc-free water.

g. Acetate buffer solution: Mix equal volumes of 2*N* sodium acetate solution and 1+7 acetic acid solution. Extract with 10-ml portions of dithizone solution I until the last extract remains green; then extract with carbon tetrachloride to remove excess dithizone.

h. Sodium thiosulfate solution: Dissolve 25 g $Na_2S_2O_3 \cdot 5H_2O$ in 100 ml zinc-free water. Purify by dithizone extraction as in ¶ 3*g* above.

i. Dithizone solution I: Dissolve 100 mg diphenylthiocarbazone* in 1 l CCl_4. Store in a brown glass-stoppered bottle in a refrigerator. If the solution is of doubtful quality or has been stored for a long time, test for deterioration as follows: Shake 10 ml with 10 ml 1+99 NH_4OH. If the lower, CCl_4, layer is only slightly yellow, the reagent is in good condition.

j. Dithizone solution II: Dilute 1 volume of dithizone solution I with 9 volumes of CCl_4. If stored in a brown glass-stoppered bottle in a refrigerator, this solution is good for several weeks.

k. Carbon tetrachloride, CCl_4. CAUTION: Carbon tetrachloride is a toxic substance. Long-continued absorption of small amounts may be hazardous. While the solvent can be absorbed through the skin, the primary danger is through inhalation of the vapor. Prepare reagents and extract standards and samples with carbon tetrachloride in a well-ventilated hood.

l. Sodium citrate solution: Dissolve 10 g $Na_3C_6H_5O_7 \cdot 2H_2O$ in 90 ml zinc-free water. Purify by dithizone extraction as in ¶ 3*g* preceding. Use this reagent in the final cleansing of glassware.

4. Procedure

a. Preparation of colorimetric standards: To a series of 125-ml Squibb separatory funnels, thoroughly cleansed as described in ¶ 2*c* above, add 0, 1.00, 2.00, 3.00, 4.00, and 5.00 ml standard zinc solution equivalent, respectively, to provide 0, 1.00, 2.00, 3.00, 4.00, and 5.00 μg Zn. Bring each volume up to 10.0 ml by adding zinc-free water. To each funnel add 5.0 ml acetate buffer and 1.0 ml sodium thiosulfate solution, and mix. The pH should be between 4 and 5.5 at this point. To each funnel add 10.0 ml dithizone solution II, stopper, and shake vigorously for 4.0 min. Let the layers separate, dry the stem of the funnel with strips of filter paper, and run the lower (CCl_4) layer into a clean *dry* absorption cell.

* Eastman No. 3092 or equivalent.

†Trade names have been identified solely to help readers and do not imply any endorsement or recommendation by the National Academy of Sciences or the National Research Council.

b. Photometric measurement: Measure either the red color of the zinc dithizonate at 535 nm, or the green color of the unreacted dithizone at 620 nm.

Set the photometer at 100% transmittance with the blank if the 535-nm wavelength is selected. If 620 nm is used, set the blank at 10.0% transmittance. Plot a calibration curve. Run a new calibration curve with each set of samples.

c. Treatment of samples: If the zinc content is not within the working range, dilute the sample with zinc-free water or concentrate it in a silica dish. If the sample has been preserved with acid, evaporate a portion to dryness in a silica dish to remove the excess acid. Do not neutralize with sodium or ammonium hydroxide because these alkalis usually contain excessive amounts of zinc. Using a pH meter and accounting for any dilution, adjust the sample to pH 2 to 3 with HCl. Transfer 10.0 ml to a separatory funnel. Complete the analysis as described in ¶ 4*a*, beginning with the words "To each funnel add 5.0 ml acetate buffer" and continuing to the end of the paragraph.

d. Visual comparison: If a photometric instrument is not available, run the samples and standards at the same time. Compare the CCl_4 layers directly in the separatory funnels if these match in size and shape; otherwise transfer to matched test tubes or nessler tubes. The range of colors obtained with various amounts of zinc are roughly these:

Zinc μg	Color
0 (blank)	green
1	blue
2	blue-violet
3	violet
4	red-violet
5	red-violet

5. Calculation

$$\text{mg/l Zn} = \frac{\mu\text{g Zn}}{\text{ml sample}}$$

6. Precision and Accuracy

A synthetic unknown sample containing 650 μg/l Zn, 500 μg/l Al, 50 μg/l Cd, 110 μg/l Cr, 470 μg/l Cu, 300 μg/l Fe, 70 μg/l Pb, 120 μg/l Mn, and 150 μg/l Ag in distilled water was analyzed in 46 laboratories by the dithizone method with a relative standard deviation of 18.2% and a relative error of 25.9%.

Dithizone Method II

1. Principle

Zinc is separated from other metals by extraction with dithizone and is then determined by measuring the color of the zinc-dithizone complex in carbon tetrachloride. Specificity in the separation is achieved by extracting from a nearly neutral solution containing bis(2-hydroxyethyl)dithiocarbamyl ion and cyanide ion, which prevents moderate concentrations of cadmium, copper, lead, and nickel from reacting with dithizone. If excessive amounts of these metals are present, follow the special procedure given in ¶ 4*b*2) below.

The color reaction is extremely sensitive; avoid introducing extraneous zinc

during the analysis. Contamination may arise from water, reagents, and glassware, such as beakers and separatory funnels, on which zinc has been adsorbed during previous use. Appreciable blanks are generally found and the analyst must satisfy himself that these blanks are representative and reproducible.

2. Apparatus

a. Colorimetric equipment: One of the following is required:

1) *Spectrophotometer*, for use at 535 nm, providing a light path of 1 cm or longer.

2) *Filter photometer*, providing a light path of 1 cm or longer and equipped with a greenish yellow filter with maximum transmittance near 535 nm.

b. Separatory funnels, 125-ml, Squibb form, with ground-glass stoppers.

3. Reagents

a. Standard zinc solution: Dissolve 1,000 g zinc metal in 10 ml 1+1 HNO_3. Dilute and boil to expel oxides of nitrogen. Transfer to a 1,000-ml volumetric flask and dilute to volume; 1.00 ml = 1.00 mg Zn.

b. Redistilled water: Distilled water redistilled in all-glass apparatus.

c. Methyl red indicator: Dissolve 0.1 g methyl red sodium salt and dilute to 100 ml with distilled water.

d. Sodium citrate solution: Dissolve 10 g $Na_3C_6H_5O_7 \cdot 2H_2O$ in 90 ml water. Shake with 10 ml dithizone solution I to remove zinc, then filter.

e. Ammonium hydroxide, NH_4OH, conc: Prepare according to directions in Section 305C.3*k*.

f. Potassium cyanide solution: Dissolve 5 g KCN in 95 ml redistilled water. (CAUTION: *Toxic—take care to avoid ingestion.*)

g. Acetic acid, conc.

h. Carbon tetrachloride, CCl_4, zinc-free. CAUTION: Carbon tetrachloride is a toxic substance. Long-continued absorption of small amounts may be hazardous. While the solvent can be absorbed through the skin, the primary danger is through inhalation of the vapor. Prepare reagents and extract standards and samples with carbon tetrachloride in a well-ventilated hood.

i. Bis (2-hydroxyethyl) dithiocarbamate solution: Dissolve 4.0 g diethanolamine and 1 ml CS_2 in 40 ml methyl alcohol. Prepare every 3 or 4 days.

j. Dithizone solution: Dilute 50 ml stock dithizone II solution (carbon tetrachloride), prepared in accordance with Section 301C.II4*b*, to 250 ml with CCl_4. Prepare fresh daily.

k. Sodium sulfide solution I: Dissolve 3.0 g $Na_2S \cdot 9H_2O$ or 1.65 g $Na_2S \cdot 3H_2O$ in 100 ml zinc-free water.

l. Sodium sulfide solution II: Prepare just before use by diluting 4 ml sodium sulfide solution I to 100 ml.

m. Nitric acid, HNO_3, 6*N*.

n. Hydrogen sulfide, H_2S.

4. Procedure

a. Preparation of calibration curve:

1) Prepare, just before use, a zinc solution containing 2.0 μg Zn/ml by diluting 5 ml standard zinc solution to 250 ml, then diluting 10 ml of the latter solution to 100 ml with redistilled water. Pipet 5.00, 10.00, 15.00, and 20.00 ml, containing 10 to 40 μg Zn, into separate 125-ml separatory funnels

and adjust the volumes to about 20 ml. Set up another funnel containing 20 ml zinc-free water as a blank.

2) Add 2 drops methyl red indicator and 2.0 ml sodium citrate solution to each funnel. If the indicator is not yellow, add conc NH_4OH a drop at a time until it just turns yellow. Add 1.0 ml potassium cyanide solution and then acetic acid, a drop at a time, until the indicator just turns a neutral peach color.

3) Extract the methyl red by shaking with 5 ml CCl_4. Discard the yellow CCl_4 layer. Add 1 ml dithiocarbamate solution. Extract with 10 ml dithizone solution, shaking for 1 min.

Draw off the CCl_4 layer into another separatory funnel and repeat the extraction with successive 5-ml portions of dithizone solution until the last one shows no change from the green dithizone color. Discard the aqueous layer.

4) Shake the combined dithizone extracts with a 10-ml portion of sodium sulfide solution II, separate the layers, and repeat the washing with further 10-ml portions of Na_2S solution until the unreacted dithizone solution has been removed completely, as shown by color of the aqueous layer, which remains colorless or very pale yellow; usually three washings are sufficient.

Remove water adhering to the stem of the funnel with a cotton swab and drain the pink CCl_4 solution into a dry 50-ml volumetric flask. Use a few milliliters of fresh CCl_4 to rinse the last droplets from the funnel and dilute to the mark with fresh CCl_4.

5) Determine the absorbance of the zinc dithizonate solutions at 535 nm, using CCl_4 as a reference. Plot an absorbance-concentration curve after subtracting the absorbance of the blank. The calibration curve is linear if monochromatic light is used.

6) Clean separatory funnels by shaking several minutes successively with HNO_3, distilled water, and finally a mixture of 5 ml sodium citrate and 5 ml dithizone, to minimize the large or erratic blanks that result from the adsorption of zinc on the glass surface. If possible, reserve separatory funnels exclusively for the zinc determination and do not use for other purposes.

b. Treatment of sample:

1) Digest sample as directed under Preliminary Treatment, Section 301C.II. Transfer a portion containing 10 to 40 μg Zn to a clean 125-ml separatory funnel and adjust the volume to about 20 ml. Determine the zinc in this solution exactly as described in the preceding procedure for preparing the calibration curve.

If more than 30 ml of dithizone solution is needed to extract the zinc completely, the portion taken contains too much zinc or the quantity of other metals that react with dithizone exceeds the amount that can be withheld by the complexing agent. In the latter case, follow the procedure in ¶ 4*b*2) below.

2) Separation of excessive amounts of cadmium, copper, and lead—When the quantity of these metals, separately or jointly, exceeds 2 mg in the portion taken, in a 100-ml beaker adjust the volume to about 20 ml. Adjust acidity to 0.4 to 0.5*N** by adding dilute HNO_3 or NH_4OH as necessary. Pass H_2S into the cold solution for 5 min. Filter off the

* The normalities of the solutions obtained in the preliminary treatment are approximately 3*N* for the HNO_3-H_2SO_4 digestion and approximately 0.8*N* for the HNO_3-$HClO_4$ digestion.

precipitated sulfides using a sintered-glass filter and wash the precipitate with two small portions of hot water. Boil the filtrate 3 to 4 min to remove H_2S, cool, transfer to a separatory funnel, and determine the zinc as described in ¶ 4*b*1) et seq.

5. Calculation

$$\text{mg/l Zn} = \frac{\mu\text{g Zn}}{\text{ml sample}} \times \frac{100}{\text{ml portion}}$$

Zincon Method*

1. General Discussion

a. Principle: Zinc forms a blue complex with 2-carboxy-2'-hydroxy-5'-sulfoformazyl benzene (zincon) in a solution buffered to pH 9.0. Other heavy metals likewise form colored complexes. Heavy metals, including zinc, are complexed by cyanide. Chloral hydrate is added specifically to free the zinc from its cyanide complex. The zinc-zincon complex is measured before other heavy metal-cyanide complexes are destroyed by chloral hydrate. Sodium ascorbate reduces the interference of manganese. The final solutions are unstable and the procedure is designed to minimize the effects of color fading.

b. Interference: The following ions interfere in concentrations exceeding those listed:

Ion	*mg/l*	Ion	*mg/l*
Cd (II)	1	Cr (III)	10
Al (III)	5	Ni (II)	20
Mn (II)	5	Cu (II)	30
Fe (III)	7	Co (II)	30
Fe (II)	9	CrO_4 (II)	50

c. Minimum detectable quantity: 1 µg Zn.

* This method, with modifications, is identical in source and substance to ASTM D1691-67.

2. Apparatus

Colorimetric equipment: One of the following is required:

a. Spectrophotometer, for measurements at 620 nm, providing a light path of 1 cm or longer.

b. Filter photometer, providing a light path of 1 cm or longer and equipped with a red filter having maximum transmittance near 620 nm. Deviation from Beer's law occurs when the filter band pass exceeds 20 nm.

3. Reagents

a. Zinc-free water, for rinsing of apparatus and preparation of solutions and dilutions. Prepare as directed in Section 323C.3*a*.

b. Stock zinc solution: Prepare as directed in Section 323C.3*b*.

c. Standard zinc solution: Dilute 10.00 ml stock zinc solution to 100 ml with zinc-free water; 1.00 ml = 10.0 µg Zn.

d. Sodium ascorbate, fine granular powder.†

e. Potassium cyanide solution: Dissolve 1.00 g KCN in 50 ml zinc-free water and dilute to 100 ml. This solution is stable for approximately 60 days.

† Hoffman-LaRoche or equivalent.

CAUTION: *Poison—potassium cyanide is extremely poisonous. Observe more than customary precautions in its handling. Never use mouth pipets to deliver volumes of cyanide solution.*

f. Buffer solution, pH 9.0: Prepare 1*N* NaOH by dissolving 40 g sodium hydroxide in 500 ml zinc-free water and diluting to 1,000 ml. Dilute 213 ml 1*N* NaOH to approximately 600 ml with zinc-free water. Dissolve 37.8 g KCl and 31.0 g H_3BO_3 in the solution and dilute to 1 l.

g. Zincon reagent: Grind the entire supply of zincon powder and make a uniform mixture. Dissolve 130 mg powdered 2-carboxy-2′-hydroxy-5′-sulfoformazyl benzene (zincon) in 100 ml methyl alcohol (methanol). Let stand overnight or use a magnetic stirrer in a closed flask to complete solution.

h. Chloral hydrate solution: Dissolve 10.0 g chloral hydrate in 50 ml zinc-free water and dilute to 100 ml. Filter if necessary.

i. Hydrochloric acid, HCl, conc.

j. Sodium hydroxide, NaOH, 6*N*.

4. Procedure

a. Preparation of colorimetric standards: To a series of thoroughly cleansed 50-ml erlenmeyer flasks, add 0, 0.25, 0.50, 1.00, 3.00, 5.00, and 7.00 ml standard zinc solution equivalent, to provide 0, 2.50, 5.00, 10.0, 30.0, 50.0, and 70.0 μg Zn, respectively. Bring each volume to 10.0 ml by adding zinc-free water. To each flask add, in sequence, mixing thoroughly after each addition, 0.5 g sodium ascorbate, 1.0 ml KCN solution, 5.0 ml buffer solution, and 3.0 ml zincon solution. Add 3.0 ml chloral hydrate solution, note the time, and mix. Transfer to the absorption cell and measure the absorbance at 620 nm *exactly* 5 min after adding the chloral hydrate solution. Use the treated blank as the reference solution for initial balancing of the photometer. For greater accuracy in the range below 10 μg Zn, prepare a separate calibration curve.

b. Treatment of samples: If dissolved zinc is to be determined, filter the sample. If total zinc is to be determined, add 1 ml conc HCl to 50 ml thoroughly mixed sample and mix well. Filter and adjust to pH 7 with 6*N* NaOH. Transfer a 10.0-ml portion of sample containing not more than 70 μg Zn to a 50-ml erlenmeyer flask. Complete the analysis as described in ¶ 4*a* above, beginning with the words "To each flask add, in sequence . . . ," and continue to the end of the paragraph.

Prepare as a reference solution a sample portion treated as above, except that 3.0 ml zinc-free water is substituted for the 3.0 ml chloral hydrate. Use this to compensate for color, turbidity, or interference not eliminated by the procedure. Prepare as nearly simultaneously as possible with the sample portion.

5. Calculation

$$\text{mg/l Zn} = \frac{\mu\text{g Zn}}{\text{ml sample}}$$

6. Precision and Accuracy

A synthetic unknown sample containing 650 μg/l Zn, 500 μg/l Al, 50 μg/l Cd, 110 μg/l Cr, 470 μg/l Cu, 300 μg/l Fe, 70 μg/l Pb, 120 μg/l Mn, and 150 μg/l Ag in distilled water was analyzed in four laboratories by the zincon method with a relative standard deviation of 13.9% and a relative error of 17.4%.

REFERENCES

1. Abbott, D. F., A. N. Exton-Smith, P. H. Millard, and J. M. Temperley. Zinc sulphate and bedsores. Brit. Med. J. 2:763, 1968. (letter)

1a. Aamodt, R., W. Rumble, S. O'Reilly, G. Johnston, and R. Henkin. Studies on the metabolism of Zn-65 in man. Fed. Proc. 34:922, 1975. (abstract)

2. Abdulla, M., and B. Haeger-Aronsen. ALA-dehydratase activation by zinc. Enzyme 12:708-710, 1971.

3. Abdulla, M., and A. Norden. Effect of food on zinc absorption. Lancet 1: 217, 1974. (letter)

4. Abernethy, R. F., and F. H. Gibson. Rare Elements in Coal. U. S. Bureau of Mines Information Circular 8163. Washington, D. C.: U. S. Department of the Interior, 1963. 69 pp.

5. Abernethy, R. F., J. M. Peter, and F. H. Gibson. Spectrochemical Analyses of Coal Ash for Trace Elements. U. S. Bureau of Mines Report of Investigations 7281. Washington, D. C.: U. S. Department of the Interior, 1969. 30 pp.

6. Addink, A. D. F. Some Aspects of Carbonic Anhydrase of Sepia officinalis (L.). Ph.D. Thesis. Utrecht, Netherlands: University of Utrecht, 1968. 161 pp.

7. Addink, H. W. H., and L. J. P. Frank. Remarks apropos of analysis of trace elements in human tissues. Cancer 12:544-551, 1959.

8. Addink, N. W. H. Subnormal level of carbonic anhydrase in blood of carcinoma patients. Nature 186:253, 1960.

9. Adelstein, S. J., and B. L. Vallee. Zinc in beef liver glutamic dehydrogenase. J. Biol. Chem. 233:589-593, 1958.

10. Affleck, R. J. Zinc poisoning in a trout hatchery. Austral. J. Marine Freshwater Res. 3:142-169, 1952.

11. Agergaard, N., and B. Palludan. Zinc metabolism in swine. VII. Alkaline phosphatase activity in plasma and tissues in relation to zinc status. Aarsberetn. Inst. Sterilitetsforsk. Kgl. Vet. Landbohoejsk. 17:47-59, 1974. (in Danish)

12. Agricola, G. (H. C. Hoover and L. H. Hoover, Translators) De Re Metallica. New York: Dover Publications, Inc., 1950. p. 354.

13. Åkeson, Å. On the zinc content of horse liver alcohol dehydrogenase. Biochem. Biophys. Res. Commun. 17:211-214, 1964.

14. Alben, A. O., J. R. Cole, and R. D. Lewis. Chemical treatment of pecan rosette. Phytopathology 22:595-601, 1932.

15. Alberts, J. C., J. A. Lang, P. S. Reyes, and G. M. Briggs. Zinc requirement of the young guinea pig. Fed. Proc. 34:906, 1975. (abstract)

16. Alexander, F. W., H. T. Delves, and H. Lay. Plasma copper and zinc in acute leukaemia. Arch. Dis. Child. 47:671, 1972. (abstract)

17. Alexander, G. V., and R. E. Nusbaum. Zinc in bone. Nature 195:903, 1962.

18. Alexander, O. R., and L. V. Taylor. Improved dithizone procedure for determination of zinc in foods. J. Assoc. Offic. Agric. Chem. 27: 324-331, 1944.

19. Alfaro, B., and F. W. Heaton. The subcellular distribution of copper, zinc and iron in liver and kidney. Changes during copper deficiency in the rat. Brit. J. Nutr. 32:435-445, 1974.

20. Allen, G. S. An outbreak of zinc poisoning in cattle. Vet. Rec. 83: 8-9, 1968.

21. Alley, M. M., D. C. Martens, M. G. Schnappinger, Jr., and G. W. Hawkins. Field calibration of soil tests for available zinc. Soil Sci. Soc. Amer. Proc. 36:621-624, 1972.

22. Altmann, H., F. Fetter, and K. Kaindl. Untersuchungen über den Einfluss un Zn-Ionen auf die m-RNS-Synthese in Chlorellazellen. Z. Naturforsch. 23B:395-396, 1968.

23. Alvares, O. F., and J. Meyer. Regional difference in parakeratotic response to mild zinc deficiency. Arch. Dermatol. 98:191-201, 1968.

24. Alvares, O. F., and J. Meyer. Thymidine uptake and cell migration in cheek epithelium of zinc-deficient rats. J. Oral Path. 2:86-94, 1973.

25. Alvares, O. F., J. Meyer, and S. J. Gerson. Activity and distribution of acid phosphatase in zinc-deficient parakeratotic rat buccal epithelium. Scand. J. Dent. Res. 81:481-488, 1973.

25a. Amador, M., M. Pena, and A. Garcia-Miranda. Low hair-zinc concentrations in acrodermatitis enteropathica. Lancet 1:1379, 1975.

26. Ambler, J. E., and J. C. Brown. Cause of differential susceptibility to zinc deficiency in two varieties of navy beans (Phaseolus vulgaris L.). Agron. J. 61:41-43, 1969.

27. Ambler, J. E., J. C. Brown, and H. G. Gauch. Effect of zinc on translocation of iron in soybean plants. Plant Physiol. 46:320-323, 1970.

28. American Conference of Governmental Industrial Hygienists. Zinc oxide fume, pp. 284-285. In Documentation of the Threshold Limit Values for Substances in the Workroom Air. (3rd ed.) Cincinnati: American Conference of Governmental Industrial Hygienists, 1971.

28a. American Bureau of Metal Statistics. Zinc, pp. 67-94. In Year Book of the American Bureau of Metal Statistics. Fifty-second Annual Issue for the Year 1972. New York: American Bureau of Metal Statistics, 1973.

28b. American Public Health Association. Subcommittee on Water Quality Control. Water Quality Standards of the United States, Territories, and the District of Columbia. Washington, D. C.: American Public Health Association, 1969. [134 pp.]

29. American Society for Testing and Materials. 1969 Book of ASTM Standards. Part 23. Water; Atmospheric Analysis. Philadelphia: American Society for Testing and Materials, 1969. 987 pp.

30. American Society for Testing and Materials. Standard method for collection and analysis of dustfall (settleable particulates), (designation ASTM D 1739-70), pp. 893-897. In 1973 Annual Book of ASTM Standards, Part 23. Water; Atmospheric Analysis. Philadelphia: American Society for Testing and Materials, 1973.

30a. American Society for Testing and Materials. Standard specification for zinc metal (slab zinc), Designation B6-70, pp. 14-17. In 1975 Annual Book of ASTM Standards. Part 8. Nonferrous Metals - Nickel, Lead, and Tin Alloys, Precious Metals, Primary Metals; Reactive Metals. Philadelphia: American Society for Testing and Materials, 1975.

31. American Society for Testing and Materials. Standard specification for zind oxide, designation D79-44 (reapproved 1974), pp. 28-29. In 1975 Annual Book of ASTM Standards. Part 28. Paint - Pigments, Resins and Polymers. Philadelphia: American Society for Testing and Materials, 1975.

31a. American Institute of Steel Construction. Specification for Structural Joints Using ASTM A325 or A490 Bolts. New York: American Institute of Steel Construction, 1976. 21 pp.

32. Ammerman, C. B., and S. M. Miller. Biological availability of minor mineral ions: A review. J. Anim. Sci. 35:681-694, 1972.

33. Armstrong, J. M., D. V. Myers, V. Verpoorte, and J. T. Edsall. Purification and properties of human erythrocyte carbonic anhydrases. J. Biol. Chem. 241:5137-5149, 1966.

34. Analytical Methods Committee: Report Prepared by the Metallic Impurities in Organic Matter Subcommittee. The determination of small amounts of zinc in organic matter by atomic-absorption spectroscopy. Analyst 98:458-460, 1973.

35. Anderson, E. A., C. E. Reinhard, and W. D. Hammel. The corrosion of zinc in various waters. J. Amer. Water Works Assoc. 26:49-60, 1934.

36. Andrási, A., and I. Fehér. Measurement of the retention and excretion of incorporated ^{65}Zn. Health Phys. 13:915-916, 1967.

37. Andreeva, T. B., and M. A. Kabanova. Determination of trace element levels in soils by atomic absorption spectrophotometry. Khim. Sel. Khoz. 6:869-873, 1968. (in Russian)

38. Andresen, E., A. Basse, E. Brummerstedt, and T. Flagstad. Zinc and the immune system in cattle. Lancet 1:839-840, 1973. (letter)

38a. Andresen, E., T. Flagstad, A. Basse, and E. Brummerstedt. Evidence of a lethal trait, A 46, in black pied Danish cattle of Friesian descent. Nord. Vet. Med. 22:473-485, 1970.

39. Anissimova, V. Experimental zinc teratomas of the testis and their transplantations. Amer. J. Cancer 36:229-232, 1939.

40. Anke, M., A. Hennig, H.-J. Schneider, H. Lüdke, W. von Gagern, and H. Schlegel. The interrelations between cadmium, zinc, copper and iron in metabolism of hens, ruminants and man, pp. 317-320. In C. F. Mills, Ed. Trace Element Metabolism in Animals. Proceedings of WAAP/IBP International Symposium, Aberdeen, Scotland, July 1969. London: E. & S. Livingston, 1970.

41. Anthony, W. L., J. M. Hsu, and F. L. Iber. Stimulation of thymidine-methyl-^{14}C oxidation in zinc-deficient rats. Fed. Proc. 34:907, 1975. (abstract)

42. Anthony, W. L., R. L. Woolsey, and J. M. Hsu. Urinary excretion of radiosulfur following taurine-^{35}S injection in zinc-deficient rats. Proc. Soc. Exp. Biol. Med. 138:989-992, 1971.

43. Antonovics, J., A. D. Bradshaw, and R. G. Turner. Heavy metal tolerance in plants. Adv. Ecol. Res. 7:1-85, 1971.

44. Apgar, J. Effect of a low zinc diet during gestation on reproduction in the rabbit. J. Anim. Sci. 33:1255-1258, 1971.

45. Apgar, J. Effect of zinc deficiency on maintenance of pregnancy in the rat. J. Nutr. 100:470-476, 1970.

46. Apgar, J. Effect of zinc deprivation from day 12, 15, or 18 of gestation on parturition in the rat. J. Nutr. 102:343-348, 1972.

47. Apgar, J. Effect of zinc repletion late in gestation on parturition in the zinc-deficient rat. J. Nutr. 103:973-981, 1973.

48. Applebury, M. L., and J. E. Coleman. Escherichia coli alkaline phosphatase. Metal binding, protein conformation, and quaternary structure. J. Biol. Chem. 244:308-318, 1969.

49. Aras, H. K., W. H. Zoller, G. E. Gordon, and G. J. Lutz. Instrumental photon activation analysis of atmospheric particulate material. Anal. Chem. 45:1481-1490, 1973.

50. Archibald, J. G. Trace elements in milk: A review -- Part II. Dairy Sci. Abstr. 20:799-812, 1958.

51. Arora, S. P., and B. S. Dhodapkar. Comparative effect of zinc oxide and zinc sulphate as dietary zinc supplements upon swine growth: A short note. Indian J. Anim. Sci. 42:525-528, 1972.

52. Arora, S. P., E. E. Hatfield, F. C. Hinds, and U. S. Garrigus. Influence of dietary zinc on the activity of blood vitamin A, alcohol dehydrogenase and carbonic anhydrase in lambs. Indian J. Anim. Sci. 43:140-144, 1973.

53. Aitchison, L. A History of Metals. Vol. II. London: MacDonald & Evans, Ltd., 1960. p. 325.

54. Athanassiadis, Y. C. Preliminary Air Pollution Survey of Zinc and Its Compounds. A Literature Review. National Air Pollution Control Administration Publication No. APTD 69-49. Raleigh, N. C.: U. S. Department of Health, Education, and Welfare, 1969. 77 pp.

55. Atkins, C. A., B. D. Patterson, and D. Graham. Plant carbonic anhydrases. I. Distribution of types among species. Plant Physiol. 50:214-217, 1972.

56. Atkins, C. A., B. D. Patterson, and D. Graham. Plant carbonic anhydrases. II. Preparation and some properties of monocotyledon and dicotyledon enzyme types. Plant Physiol. 50:218-223, 1972.

57. Atkinson, J., P. Vohra, and F. H. Kratzer. The effect of available dietary zinc on the utilization of protein by the chick and Japanese quail. Brit. J. Nutr. 27:461-466, 1972.

58. Auerbach, S. Zinc content of plasma, blood, and erythrocytes in normal subjects and in patients with Hodgkin's disease and various hematologic disorders. J. Lab. Clin. Med. 65:628-637, 1965.

59. Auld, D. S., H. Kawaguchi, D. M. Livingston, and B. L. Vallee. RNA-dependent DNA polymerase (reverse transcriptase) from avian myeloblastosis virus: A zinc metalloenzyme. Proc. Nat. Acad. Sci. U.S.A. 71:2091-2095, 1974.

59a. Auld, D. S., H. Kawaguchi, D. M. Livingston, and B. L. Vallee. Zinc reverse transcriptases from mammalian RNA type C viruses. Biochem. Biophys. Res. Commun. 62:296-302, 1975.

59b. Auld, D. S., H. Kawaguchi, D. M. Livingston, and B. L. Vallee. Reverse transcriptase from avian myeloblastosis virus: A zinc metalloenzyme. Biochem. Biophys. Res. Commun. 57:967-972, 1974.

60. Axelsson, B., and M. Piscator. Renal damage after prolonged exposure to cadmium. An experimental study. Arch. Environ. Health 12:360-373, 1966.

61. Babatunde, G. M., and B. L. Fetuga. Zinc storage in the tissues and organs of pigs fed graded levels of zinc in the tropics. Anim. Prod. 15:21-28, 1972.

62. Bachmann, R. W. Zinc-65 in studies of the freshwater zinc cycle, pp. 485-496. In V. Schultz and A. W. Klement, Eds. Radioecology. New York: Reinhold Publishing Corporation, 1963.

63. Bachmann, R. W., and E. P. Odum. Uptake of Zn^{65} and primary productivity in marine benthic algae. Limnol. Oceanogr. 5:349-355, 1960.

64. Bagg, H. J. Experimental production of teratoma testis in the fowl. Amer. J. Cancer 26:69-84, 1936.

65. Baker, D. H., A. J. Kleiss, B. G. Harmon, and A. H. Jensen. Levels of dietary zinc for pregnant gilts. J. Anim. Sci. 35:1101, 1972. (abstract)

66. Bakker, A. Über Kohlensäureanhydrase in normalen und kataraktösen Linsen. Albrecht von Graefes Arch. Ophthalmol. 140:543-552, 1939.

67. Ball, I. R. The relative susceptibilities of some species of fresh-water fish to poisons. —I. Ammonia. Water Res. 1:767-775, 1967.

68. Ballou, J. E., and R. C. Thompson. Metabolism of zinc-65 in the rat. Consideration of permissible exposure limits. Health Phys. 6:6-18, 1961.

69. Banat, K., U. Förstner, and G. Müller. Schwermetalle in Sedimenten von Donau, Rhein, Ems, Weser, und Elbe im Bereich der Bundesrepublik Deutschland. Naturwissenschaften 59:525-528, 1972.

70. Banis, R. J., W. G. Pond, E. F. Walker, Jr., and J. R. O'Connor. Dietary cadmium, iron, and zinc interactions in the growing rat. Proc. Soc. Exp. Biol. Med. 130:802-806, 1969.

71. Baptist, J. P., D. E. Hoss, and C. W. Lewis. Retention of ^{51}Cr, ^{59}Fe, ^{60}Co, ^{65}Zn, ^{85}Sr, ^{95}Nb, ^{141m}In and ^{131}I by the Atlantic croaker (Micropogon undulatus). Health Phys. 18:141-148, 1970.

72. Baptist, J. P., and C. W. Lewis. Transfer of ^{65}Zn and ^{51}Cr through an estuarine food chain, pp. 420-430. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

73. Barber, S. A. A diffusion and mass-flow concept of soil nutrient availability. Soil Sci. 93:39-49, 1962.

74. Barber, S. A., J. M. Walker, and E. H. Vasey. Mechanisms for the movement of plant nutrients from the soil and fertilizer to the plant root. J. Agric. Food Chem. 11:204-207, 1963.

75. Barbiroli, B., M. S. Moruzzi. B. Tadolini, and M. G. Monti. Ornithine decarboxylase activity in regenerating liver from rats adapted to controlled feeding schedules. J. Nutr. 105:408-412, 1975.

75a. Barcia, P. J. Lack of acceleration of healing with zinc sulfate. Ann. Surg. 172:1048-1050, 1970.

76. Bargetzi, J.-P., K. S. V. Kumar, D. J. Cox, K. A. Walsh, and H. Neurath. The amino acid composition of bovine pancreatic carboxypeptidase A. Biochemistry 2:1468-1474, 1963.

77. Barnes, H. The determination of zinc by dithizone. Analyst 76:220-223, 1951.

77a. Barnes, P. M., and E. J. Moynahan. Zinc deficiency in acrodermatitis enteropathica: Multiple dietary intolerance treated with synthetic diet. Proc. Roy. Soc. Med. 66:327-329, 1973.

78. Barnette, R. M., J. P. Camp, J. D. Warner, and O. E. Gall. The Use of Zinc Sulphate Under Corn and Other Field Crops. Bulletin 292. Gainesville: University of Florida, Agricultural Experiment Station, 1936. 51 pp.

79. Barney, G. H., M. P. Macapinlac, W. N. Pearson, and W. J. Darby. Parakeratosis of the tongue -- a unique histopathological lesion in the zinc-deficient squirrel monkey. J. Nutr. 93:511-517, 1967.

80. Barney, G. H., M. C. Orgebin-Crist, and M. P. Macapinlac. Genesis of esophageal parakeratosis and histological changes in the testes of the zinc-deficient rat and their reversal by zinc repletion. J. Nutr. 95:526-534, 1968.

81. Barr, D. H., and J. W. Harris. Growth of the P388 leukemia as an ascites tumor in zinc-deficient mice. Proc. Soc. Exp. Biol. Med. 144:284-287, 1973.

82. Barrett, G. V., and R. H. Franke. "Psychogenic" death: A reappraisal. Science 167:304-306, 1970.

83. Bartow, E., and O. M. Weigle. Zinc in water supplies. Ind. Eng. Chem. 24:463-465, 1932.

84. Batchelor, R. P., J. W. Fehnel, R. M. Thomson, and K. R. Drinker. A classical and laboratory investigation of effect of metallic zinc, of zinc oxide and of zinc sulphide upon health of workmen. J. Ind. Hyg. 8:322-363, 1926.

85. Bauer, A. Considerations in the development of soil tests for "available" zinc. Commun. Soil Sci. Plant Anal. 2:161-194, 1971.

86. Bauer, A., and W. L. Lindsay. The effect of soil temperature on the availability of indigenous soil zinc. Soil Sci. Soc. Amer. Proc. 29:413-416, 1965.

87. Baughman, N. M. The Effect of Organic Matter on the Retention of Zinc by the Soil. Ph.D. Thesis. Lafayette, Ind.: Purdue University, 1956. 96 pp.

88. Baumeister, W. Schwermetall-Pflanzengesellschaften und Zinkresistenz einiger Schwermetallpflanzen. Angew. Bot. 40:185-204, 1967.

89. Baumslag, N., D. Yeager, L. Levin, and H. G. Petering. Trace metal content of maternal and neonate hair. Zinc, copper, iron, and lead. Arch. Environ. Health 29:186-191, 1974.

89a. Baxter, D. J., and W. O. Smith. Uptake of radioactive zinc and manganese in damaged rat liver. Proc. Soc. Exp. Biol. Med. 109:287-288, 1962.

90. Beauchamp, E. G., and G. Lean. Evaluation of surfactants for zinc absorption by soybean leaf tissues. Commun. Soil Sci. Plant Anal. 4:1-7, 1973.

91. Becker, R. O., J. A. Spadaro, and E. W. Berg. The trace elements in human bone. J. Bone Joint Surg. 50A:326-334, 1968.

92. Becker, W. M., and W. G. Hoekstra. Effect of vitamin D on ^{65}Zn absorption, distribution and turnover in rats. J. Nutr. 90:301-309, 1966.

93. Becking, G. C., and A. B. Morrison. Hepatic drug metabolism in zinc-deficient rats. Biochem. Pharmacol. 19:895-902, 1970.

94. Beeckmans, J. M., and J. R. Brown. Toxicity of catalytically active zinc oxides. Arch. Environ. Health 7:346-350, 1963.

94a. Beisel, W. R., and R. S. Pekarek. Acute stress and trace element metabolism. Int. Rev. Neurobiol. Suppl. 1:53-82, 1972.

94b. Beisel, W. R., R. S. Pekarek, D. van Ormer, and R. W. Wannemacher, Jr. Influence of acute infection on the metabolism of zinc and other trace elements. Psychopharmacol. Bull. 7(3):34-35, 1971.

94c. Beisel, S. R., R. S. Pekarek, and R. W. Wannemacher, Jr. The impact of infectious disease on trace-element metabolism of the host, pp. 217-240. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals--2. Proceedings of 2nd International Symposium, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

95. Berfenstam, R. Studies on blood zinc. A clinical and experimental investigation into the zinc content of plasma and blood corpuscles with special reference to infancy. Acta Paediatr. 41(Suppl. 87):1-105, 1952.

96. Berg, L. R., and R. D. Martinson. Effect of diet composition on the toxicity of zinc for the chick. Poult. Sci. 51:1690-1694, 1972.

97. Berger, K. C. Introductory Soils. New York: The MacMillan Co., 1965. 371 pp.

98. Berger, N. A., and A. M. Skinner. Characterization of lymphocyte transformation induced by zinc ions. J. Cell Biol. 61:45-55, 1974.

99. Bergman, B. The distribution and concentration of zinc and the effect of zinc deficiency in the mammalian body. Some experiments in mice and rats with special reference to mandibular condyle and some other skeletal tissues. Odontol. Revy 21(Suppl. 20), 1970. 54 pp.

100. Bergman, B. The zinc concentration in hard and soft tissues of the rat. The influence of zinc deficient feeding. Acta Odontol. Scand. 28: 425-440, 1970.

101. Bergman, B., U. Friberg, S. Lohmander, and T. Öberg. Morphologic and autoradiographic observations on the effect of zinc deficiency on endochondral growth sites in the white rat. Odontol. Revy 21: 379-399, 1970.

102. Bergman, B., U. Friberg, S. Lohmander, and T. Öberg. The importance of zinc to cell proliferation in endochondral growth sites in the white rat. Scand. J. Dent. Res. 80:486-492, 1972.

103. Bergman, B., R. Sjöström, and K. R. Wing. Variation with the age of tissue zinc concentrations in albino rats determined by atomic absorption spectrophotometry. Acta Physiol. Scand. 92:440-450, 1974.

104. Bergman, B., and K. R. Wing. Turnover of ^{65}Zn in rats fed a zinc-deficient diet. Acta Physiol. Scand. 92:451-464, 1974.

105. Bernhard, M., and A. Zattera. A comparison between the uptake of radioactive and stable zinc by a marine unicellular alga, pp. 389-398. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

106. Berrow, M. L., and J. Webber. Trace elements in sewage sludges. J. Sci. Food Agric. 23:93-100, 1972.

107. Bertine, K. K., and E. D. Goldberg. Fossil fuel combustion and the major sedimentary cycle. Science 173:233-235, 1971.

108. Bertrand, D., and A. de Wolf. Premières recherches sur le rôle de l'oligoélement zinc chez l'Aspergillus niger. Bull. Soc. Chim. Biol. 41:545-554, 1959.

108a. Bertrand, G., and R. C. Bhattacherjee. L'action combinée du zinc et des vitamins dans l'alimentation des animaux. C. R. Acad. Sci. (Paris) 198:1823-1827, 1934.

109. Bertrand, G., and R. Vladesco. Intervention probable du zinc dans les phénomènes de fécondation chez les animaux vertébrés. C. R. Acad. Sci. D (Paris) 173:176-179, 1921.

110. Besch, K. W., and P. Roberts-Pichette. Effects of mining pollution on vascular plants in the northwest Miramichi River system. Can. J. Bot. 48:1647-1656, 1970.

111. Biddulph, O. Translocation of radioactive mineral nutrients in plants, pp. 48-58. In A Conference on the Use of Isotopes in Plant and Animal Research. Held on June 12-14, 1952. Report No. 4 of Agricultural Experiment Station, Kansas State College of Agriculture and Applied Science, Manhattan, Kansas. Washington, D. C.: U. S. Atomic Energy Commission, 1953.

111a. Bierenbaum, M. L., A. I. Fleischman, J. P. Dunn, T. Hayton, D. C. Pattison, and P. B. Watson. Serum parameters in hard and soft water communities. Amer. J. Public Health 63:169-173, 1973.

112. Biesinger, K. E., and G. M. Christensen. Effects of various metals on survival, growth, reproduction, and metabolism of Daphnia magna. J. Fish. Res. Board Can. 29:1691-1700, 1972.

113. Bingham, F. T., and M. J. Garber. Solubility and availability of micronutrients in relation to phosphorous fertilization. Soil Sci. Soc. Amer. Proc. 24:209-213, 1960.

114. Birks, L. S., and J. V. Gilfrich. Development of X-ray Fluorescence Spectroscopy in Elemental Analysis of Particulate Matter. Phase II. Evaluation of Commercial Multiple Crystal Spectrometer Instruments. EPA 650/12-73-006. Washington, D. C.: Naval Research Laboratory, 1973. 7 pp.

115. Birks, L. S., J. V. Gilfrich, and P. G. Burkhalter. Development of X-ray Fluorescence Spectroscopy for Elemental Analysis of Particulate Matter in the Atmosphere and in Source Emission. EPA-R2-72-063. Washington, D. C.: Naval Research Laboratory, 1972. 43 pp.

116. Bischoff, F. Factors influencing the augmentation effects produced by zinc or copper when mixed with gonadotropic extracts. Amer. J. Physiol. 121:765-770, 1938.

117. Bischoff, F. Histone combinations of the protein hormones. Amer. J. Physiol. 117:182-187, 1936.

118. Bischoff, F., and M. L. Long. The local influence of zinc on the development of thymomas in Marsh-Buffalo mice. Amer. J. Cancer 38:404-405, 1940.

119. Bischoff, F., and M. L. Long. The local effect of zinc on the development of the Marsh-Buffalo adenocarcinoma. Amer. J. Cancer 37:531-535, 1939.

120. Bishop, R. F., and C. R. MacEachern. The zinc status of some Nova Scotia soils and crops. Commun. Soil Sci. Plant Anal. 4:41-50, 1973.

121. Bishop, W. H., F. A. Quiocho, and F. M. Richards. The removal and exchange of metal ions in cross-linked crystals of carboxypeptidase-A. Biochemistry 5:4077-4087, 1966.

122. Bitar, K., and J. G. Reinhold. Phytase and alkaline phosphatase activities in intestinal mucosae of rat, chicken, calf, and man. Biochim. Biophys. Acta 268:442-452, 1972.

122a. Black, O., Jr., and P. D. Webster. III. Nutritional and hormonal effects on RNA polymerase enzyme activities in pancreas. Amer. J. Physiol. 227:1276-1280, 1974.

122b. Black, O., Jr., and P. D. Webster. Protein synthesis in pancreas of fasted pigeons. J. Cell Biol. 57:1-8, 1973.

123. Black, W. A. P., and R. L. Mitchell. Trace elements in the common brown algae and sea water. J. Marine Biol. Assoc. U. K. 30:575-584, 1952.

123a. Blakeslee, P. A. Monitoring considerations for municipal wastewater effluent and sludge application to the land, pp. 183-198. In Proceedings of the Joint Conference on Recycling Municipal Sludges and Effluents on Land. Champaign, Illinois, July 9-13, 1973. Washington, D. C.: National Association of State Universities and Land Grant Colleges, 1973.

124. Blessings of zinc. Food Cosmet. Toxicol. 10:578-583, 1972.

125. Blincoe, C., and T. L. Lambert. Micronutrient trace element composition of crested wheatgrass. J. Range Manage. 25:128-130, 1972.

126. Bliss, A. F. The equilibrium between vitamin A alcohol and aldehyde in the presence of alcohol dehydrogenase. Arch. Biochem. Biophys. 31: 197-204, 1951.

127. Blomfield, J., J. McPherson, and C. R. P. George. Active uptake of copper and zinc during haemodialysis. Brit. Med. J. 2:141-145, 1969.

128. Boawn, L. C. Comparison of zinc sulfate and zinc EDTA as zinc fertilizer sources. Soil Sci. Soc. Amer. Proc. 37:111-115, 1973.

129. Boawn, L. C. Residual availability of fertilizer zinc. Soil Sci. Soc. Amer. Proc. 38:800-803, 1974.

130. Boawn, L. C. Zinc accumulation characteristeics of some leafy vegetables. Commun. Soil Sci. Plant Anal. 2:31-36, 1971.

131. Boawn, L. C., and J. G. Brown. Further evidence for a P-Zn imbalance in plants. Soil Sci. Soc. Amer. Proc. 32:94-97, 1968.

132. Boawn, L. C., C. E. Nelson, F. C. Viets, Jr., and C. L. Crawford. Nitrogen Carrier and Nitrogen Rate Influence on Soil Properties and Nutrient Uptake by Crops. Washington Agricultural Experiment Station Bulletin 614. Pullman: Washington State University. Institute of Agricultural Sciences, 1960. 24 pp.

133. Boawn, L. C., and P. E. Rasmussen. Crop response to excessive zinc fertilization of alkaline soil. Agron. J. 63:874-876, 1971.

134. Boawn, L. C., P. E. Rasmussen, and J. W. Brown. Relationship between tissue zinc levels and maturity period of field beans. Agron. J. 61:49-51, 1969.

135. Boawn, L. C., F. G. Viets, Jr., C. L. Crawford, and J. L. Nelson. Effect of nitrogen carrier, nitrogen rate, zinc rate and soil pH on zinc uptake by sorghum, potatoes, and sugar beets. Soil Sci. 90:329-337, 1960.

136. Bodansky, M. Biochemical studies on marine organisms. II. The occurrence of zinc. J. Biol. Chem. 44:399-407, 1920.

137. Bodart, F., G. Deconninck, J. Hontoy, and S. Wilk. Filter paper analysis by fluorescence measurements. Radiochem. Radioanal. Lett. 13:161-167, 1973.

138. Boddy, K., B. W. East, P. C. King, R. W. Simpson, and R. Scott. Preliminary studies on zinc metabolism in carcinoma of the prostate gland. Brit. J. Urol. 42:475-480, 1970.

139. Boquist, L., and Å. Lernmark. Effects on the endocrine pancreas in Chinese hamsters fed zinc deficient diets. Acta Path. Microbiol. Scand. 76:215-227, 1969.

140. Borisova, M. A. The indices of ceruloplasmin activity, the blood content of copper and zinc in patients with typhoid and dysentery in levomycetin therapy. Sov. Med. 29(1):59-63, 1966. (in Russian, summary in English)

140a. Boström, K., and L. Andersson. Creatine phosphokinase relative to acid phosphatase, lactase dehydrogenase, zinc in fructrose in human semen with special reference to chronic prostatitis. Scand. J. Urol. Nephrol. 5:123-132, 1971.

141. Böttger, P. Zinkbestimmungen in menschichem Sperma. Med. Welt. 24: 374-376, 1973.

142. Bourne, G. H. Phosphatase and calcification, pp. 79-120. In G. H. Bourne, Ed. The Biochemistry and Physiology of Bone. Vol. 2. Physiology and Pathology. (2nd ed.) New York: Academic Press, 1972.

143. Bournsnell, J. C., S. Baronos, P. A. Briggs, and E. J. Butler. The concentrations of zinc in boar seminal plasma and vesicular secretion in relation to those of nitrogenous substances, nitrate, galactose and fructose. J. Reprod. Fertil. 29:215-227, 1972.

144. Bournsnell, J. C., P. A. Briggs, U. Lavon, and E. J. Butler. The association of zinc with some components of boar vesicular secretion and seminal plasma. I. Gel-filtration and dialysis studies. J. Reprod. Fertil. 34:57-71, 1973.

145. Bournsnell, J. C. P. A. Briggs, U. Lavon, and E. J. Butler. The association of zinc with some components of boar vesicular secretion and seminal plasma. II. Ultrafiltration and ethanol-precipitation studies. J. Reprod. Fertil. 34:73-77, 1973.

146. Bowen, H. J. M. Trace Elements in Biochemistry. New York: Academic Press, 1966. 241 pp.

147. Bowen, J. E. Absorption of copper, zinc, and manganese by sugarcane leaf tissue. Plant Physiol. 44:255-261, 1969.

148. Bowen, J. E. Kinetics of zinc absorption by excised roots of two sugarcane clones. Plant Soil 39:125-129, 1973.

148a. Bowman, H. R., J. G. Conway, and F. Asaro. Atmospheric lead and bromine concentration in Berkeley, Calif. (1963-70). Environ. Sci. Technol. 6:558-562, 1972.

149. Boyd, C. E. Some aspects of aquatic plant ecology, pp. 114-129. In Reservoir Fishery Resources. Symposium Presented by the Reservoir Committee of the Southern Division, American Fisheries Society at University of Georgia, Athens, April 5-7, 1967. Washington, D. C.: American Fisheries Society, 1968.

149a. Boyett, J. D., and J. F. Sullivan. Zinc and collagen content of cirrhotic liver. Amer. J. Dig. Dis. 15:797-802, 1970.

150. Boyd, E. M., and K. J. Clark. The prolongation by zinc salts of a water balance reaction of posterior hypophyseal extract. Amer. J. Med. Sci. 198:171-177, 1939.

150a. Bradfield, R. B., T. Yee, and J. M. Baertl. Hair zinc levels of Andean Indian children during protein-caloric malnutrition. Amer. J. Clin. Nutr. 22:1349-1353, 1969.

151. Brand-Auraban, A., L. Kopito, and H. Schwachman. Chemical analysis of some inorganic elements in cerumen from patients with cystic fibrosis. J. Invest. Derm. 58:14-15, 1972.

152. Brech, F. Comparison of optical emission and atomic absorption methods for the analyses of plant tissues. J. Assoc. Off. Anal. Chem. 51:132-136, 1968.

153. Delete 153--use 1743a

154. Bremner, I. The nature of trace element binding in herbage and gut contents, pp. 366-369. In C. F. Mills, Ed. Trace Element Metabolism in Animals. Proceedings of WAAP/IBP International Symposium, Aberdeen, Scotland, July 1969. London: E. & S. Livingston, 1970.

155. Bremner, I. Zinc, copper and manganese in the alimentary tract of sheep. Brit. J. Nutr. 24:769-783, 1970.

155a. Bremner, I., and N. T. Davies. Studies on the appearance of a copper-binding protein in rat liver. Biochem. Soc. Trans. 2:425-427, 1974.

156. Bremner, I., N. T. Davies, and C. F. Mills. The effect of zinc deficiency and food restriction on hepatic zinc proteins in the rat. Biochem. Soc. Trans. 1:982-985, 1973.

156a. Bremner, I., and R. B. Marshall. Hepatic copper- and zinc-binding proteins in ruminants. 1. Distribution of Cu and Zn among soluble proteins of livers varying Cu and Zn content. Brit. J. Nutr. 32: 283-291, 1974.

156b. Bremner, I., and R. B. Marshall. Hepatic copper- and zinc-binding proteins in ruminants. 2. Relationship between Cu and Zn concentrations and the occurrence of a metallothionein-like function. Brit. J. Nutr. 32: 293-300, 1974.

157. Brenchley, W. E. Inorganic Plant Poisons and Stimulants. Cambridge: University Press, 1914. 110 pp.

158. Breslow, E., and A. W. Girotti. The interaction of ribonuclease with metal ions. I. Studies of cupric and zinc ions and the effect of cytidylic acid. J. Biol. Chem. 241:5651-5660, 1966.

158a. Brewer, G. J., S. Dash, and F. J. Oelshlegel, Jr. The effect of zinc on hemoglobin binding by red blood cell membranes. J. Clin. Invest. 53: 11a, 1974. (abstract)

159. Brierley, G. P. Ion transport by heart mitochondria VII. Activation of the energy-linked accumulation of Mg^{++} by Zn^{++} and other cations. J. Biol. Chem. 242:1115-1122, 1967.

160. Brierley, G. P., and R. N. Bhattacharyya. Activation of Mg^{++} accumulation in isolated heart mitochondria by Zn^{++} and by p-chloromercuribenzene sulfonate. Biochem. Biophys. Res. Commun. 23:647-651, 1966.

161. Brierley, G. P., R. N. Bhattacharyya, and J. G. Walker. Induction of K^+ transport in isolated heart mitochondria by zinc ions. Biochem. Biophys. Res. Commun. 24:269-273, 1966.

162. Brierley, G. P., W. E. Jacobus, and G. R. Hunter. Ion transport by heart mitochondria. VIII. Activation of the adenosine triphosphate-supported accumulation of Mg^{++} by Zn^{++} and by p-chloromercuriphenylsulfonate. J. Biol. Chem. 242:2192-2198, 1967.

163. Brierly, G. P., and V. A. Knight. Ion transport by heart mitochondria. X. The uptake and the release of Zn^{2+} and its relation to the energy-linked accumulation of magnesium. Biochemistry 6:3892-3901, 1967.

164. Brierley, G. P., and C. T. Settlemire. Ion transport by heart mitochrondria. IX. Induction of the energy-linked uptake of K^+ by zinc ion. J. Biol. Chem. 242:4324-4328, 1967.

165. Brill, A. S., and J. H. Venable, Jr. Effects of site symmetry and sequential metal binding upon protein titration (zinc insulin). J. Amer. Chem. Soc. 89:3622-3626, 1967.

166. Broda, E., H. Desser, and G. Findenegg. Wirkung von Dinitrophenol, Azid und Anaerobiose auf die Zinkaufnahme durch Algen. Naturwissenschaften 51:361-362, 1964.

167. Brodie, K. G., and J. P. Matoušek. Determination of cadmium in air by non-flame atomic absorption spectrometry. Anal. Chim. Acta 69:200-202, 1974.

168. Brooks, R. R., and M. G. Rumsby. The biogeochemistry of trace element uptake by some New Zealand bivalves. Limnol. Oceanogr. 10:521-527, 1965.

169. Brown, A. L., B. A. Krantz, and J. L. Eddings. Zinc-phosphorous interactions as measured by plant response and soil analysis. Soil Sci. 110:415-420, 1970.

170. Brown, J. C. Iron chlorosis in plants. Adv. Agron. 13:329-369, 1961.

171. Brown, J. C., J. E. Ambler, R. L. Chaney, and C. D. Foy. Differential responses of plant genotypes to micronutrients, pp. 389-418. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

172. Brown, R., M. L. Jacobs, and H. E. Taylor. A survey of the most recent applications of spark source mass spectrometry. Amer. Lab. 4(11): 29-40, 1972.

173. Brown, M. A., J. V. Thom, G. L. Orth, P. Cova, and J. Juarez. Food poisoning involving zinc contamination. Arch. Environ. Health 8:657-660, 1964.

174. Brown, J. C., and L. O. Tiffin. Chelates for micronutrients. Properties of chelates and their use in crop production. J. Agric. Food Chem. 10:192-195, 1962.

175. Brown, J. C., L. O. Tiffin, and R. S. Holmes. Competition between chelating agents and roots as factor affecting absorption of iron and other ions by plant species. Plant Physiol. 35:878-886, 1960.

176. Brown, R., and P. G. T. Vossen. The determination of inorganic pollutants of air and water by spark source mass spectrometry, pp. 427-431. In B. Westley, Ed. Proceedings of the International Symposium on Identification and Measurement of Environmental Pollutants. Ottawa, Ontario, Canada, June 14-17, 1971. Ottawa: National Research Council of Canada, 1971.

177. Browne, R. C. Zinc, pp. 86-87. In The Chemistry and Therapy of Industrial Diseases. Springfield, Ill.: Charles C Thomas, 1966.

178. Brudevold, F., L. T. Steadman, M. A. Spinelli, B. H. Amdur, and P. Grǿn. A study of zinc in human teeth. Arch. Oral Biol. 8:135-144, 1963.

179. Brummerstedt, E., T. Flagstad, A. Basse, and E. Andresen. The effect of zinc on calves with hereditary thymus hypoplasia (lethal trait A 46). Acta Path. Microbiol. Scand. A 79:686-687, 1971.

180. Brundell, J., S. O. Falkbring, and P. O. Nyman. Carbonic anhydrase from Neisseria sicca strain 6021 II. Properties of the purified enzyme. Biochim. Biophys. Acta 284:311-323, 1972.

181. Brune, D., K. Samsahl, and P. O. Wester. Determination of elements in milli-, micro- and submicrogram quantities in human whole blood by neutron activation analysis. Atompraxis 9:368-373, 1963.

182. Brungs, W. A. Chronic toxicity of zinc to the fathead minnow, Pimephales promelas Rafinesque. Trans. Amer. Fish. Soc. 98:272-279, 1969.

183. Brungs, W. A., Jr. Distribution of Cobalt 60, Zinc 65, Strontium 85, and Cesium 137 in a Freshwater Pond. Public Health Service Publication No. 999-RH-24. Washington, D. C.: U. S. Government Printing Office, 1967. 52 pp.

184. Brunner, E., and U. Brunner-Frühwald. Über eine Frühdiagnose des Karzinoms durch Zinknachweis im Leukozyten. Wien. Klin. Wochenschr. 77:979-980, 1965. (abstract)

185. Brunner, E., and U. Frühwald. Die Bestimmung des Zinkgehaltes der Leukozyten als Möglichkeit der Frühdiagnose des Karzinoms. Wien. Klin. Wochenschr. 78:33-34, 1966.

186. Bryan, G. W. Concentrations of zinc and copper in the tissues of decapod crustaceans. J. Marine Biol. Assoc. U. K. 48:303-321, 1968.

187. Bryan, G. W. The absorption of zinc and other metals by the brown seaweed Laminaria digitata. J. Marine Biol. Assoc. U. K. 49:225-243, 1969.

188. Bryan, G. W. The effects of heavy metals (other than mercury) on marine and estuarine organisms. Proc. Roy. Soc. London B 177:389-410, 1971.

189. Bryan, G. W. The metabolism of Zn and ^{65}Zn in crabs, lobsters, and fresh-water crayfish, pp. 1005-1016. In B. Åberg and F. P. Hungate, Eds. Radioecological Concentration Processes. Proceedings of an International Symposium held in Stockholm 25-29 April, 1966. New York: Pergamon, 1967.

190. Bryan, G. W. The occurrence and seasonal variation of trace metals in the scallops Pecten maximus (L.) and Chlamys opercularis (L.). J. Marine Biol. Assoc. U. K. 53:145-166, 1973.

191. Bryan, G. W. Zinc concentrations of fast and slow contracting muscles in the lobster. Nature 213:1043-1044, 1967.

192. Bryan, G. W. Zinc regulation in the freshwater crayfish (including some comparative copper analyses). J. Exp. Biol. 46:281-296, 1967.

193. Bryan, G. W. Zinc regulation in the lobster Homarus vulgaris. I. Tissue zinc and copper concentrations. J. Marine Biol. Assoc. U. K. 44:549-563, 1964.

194. Bryan, G. W., and L. G. Hummerstone. Adaptation of the polychaete Nereis diversicolor to estuarine sediments containing high concentrations of zinc and cadmium. J. Marine Biol. Assoc. U. K. 53:839-857, 1973.

195. Bryan, G. W., and L. G. Hummerstone. Adaptations of the polychaete Nereis diversicolor to estuarine sediments containing high concentrations of heavy metals. I. General observations and adaptation to copper. J. Marine Biol. Assoc. U. K. 51:845-863, 1971.

196. Bryan, G. W., and L. G. Hummerstone. Brown seaweed as an indicator of heavy metals in estuaries in South-West England. J. Marine Biol. Assoc. U. K. 53:705-720, 1973.

197. Bryan, G. W., A. Preston, and W. L. Templeton. Accumulation of radionuclides by aquatic organisms of economic importance in the United Kingdom, pp. 623-637. In Disposal of Radioactive Wastes into Seas, Oceans and Surface Waters. Proceedings of a Symposium, Vienna, 16-20 May 1966. Vienna: International Atomic Energy Agency, 1966.

198. Buchauer, M. J. Contamination of soil and vegetation near a zinc smelter by zinc, cadmium, copper, and lead. Environ. Sci. Technol. 7:131-135, 1973.

198a. Buck, W. R., III., and W. A. Van Engel. A Study of Zinc Anodes on Crab Pots. Summary Report to the American Zinc Institute. Richmond: Virginia Institute for Scientific Research, 1960. 5 pp.

199. Buerk, C. A., M. G. Chandy, E. Pearson, A. MacAuly, and H. S. Soroff. Zinc deficiency: Effect on healing and metabolism in man. Surg. Forum 24: 101-103, 1973.

200. Bukovac, M. J., and A. J. Riga. Redistribution of cotyledonary phosphorous, calcium and zinc during germination and early seedling development of Phaseolus vulgaris L, pp. 280-285. In XVIth International Horticultural Congress - 1962. Brussels, Belgium, Aug. 31-Sept. 8. Vol. 2. Gembloux, Belgium: J. Duculot, S. A., 1963.

201. Bunn, C. R., and G. Matrone. In vivo interactions of cadmium, copper, zinc, and iron in the mouse and rat. J. Nutr. 90:395-399, 1966.

201a. Burch. R. E., H. K. J. Hahn, and J. F. Sullivan. Newer aspects of the roles of zinc, manganese, and copper in human nutrition. Clin. Chem. 21:501-520, 1975.

201b. Burch, R. E., R. V. Williams, H. K. J. Hahn, M. M. Jetton, and J. F. Sullivan. Serum and tissue enzyme activity and trace-element content in response to zinc deficiency in the pig. Clin. Chem. 21:568-577, 1975.

202. Burke, A. D., F. Woodson, and V. G. Heller. The possible toxicity of buttermilk soured in zinc containers. J. Dairy Sci. 11:79-88, 1928.

203. Burkitt, A., P. Lester, and G. Nickless. Distribution of heavy metals in the vicinity of an industrial complex. Nature 238:327-328, 1972.

204. Burr, R. G. Blood zinc in the spinal patient. J. Clin. Path. 26:773-775, 1973.

205. Burr, R. G. Plasma-zinc levels. Lancet 1:879, 1974. (letter)

206. Burrell, D. C. Flame spectrophotometric trace analysis, pp. 477-534. In M. Zief and R. Speights, Eds. Ultrapurity: Methods and Techniques. New York: Marcel Dekker, Inc., 1972.

207. Burstein, A. Brass founders ague. J. Ind. Hyg. (Suppl.) 8:110, 1926. (abstract)

208. Burton, D. T., A. H. Jones, and J. Cairns, Jr. Acute zinc toxicity to rainbow trout (Salmo gairdneri): Confirmation of the hypothesis that death is related to tissue hypoxia. J. Fish. Res. Board Can. 29:1463-1466, 1972.

208a. Burton, J. L., and S. K. Goolamali. Zinc and sebum excretion. Lancet 1: 1448, 1973. (letter)

209. Burvill, G. H. Plant nutrition in Western Australia. J. Dept. Agric. West. Austral. Series 4. 6:353-371, 1965.

210. Bushmanov, B. S., and A. M. Rakhanov. Clinical signs of zinc phosphide poisoning of animals (Indian). Veterinarie 44(12):63-64, 1967. (in Russian)

211. Deleted.

212. Butler, P. C., and R. H. Bray. Effect of the zinc chelate of ethylenediaminetetraacetic acid on plant uptake of zinc and other heavy metals. Soil Sci. Soc. Amer. Proc. 20:348-351, 1956.

213. Butt, E. M., R. E. Nusbaum, T. C. Gilmour, S. L. Didio, and S. Mariano. Trace metal levels in human serum and blood. Arch. Environ. Health 8:52-57, 1964.

214. Byvoet, P., and A. Gotti. Isolation and properties of carbonic anhydrase from dog kidney and erythrocytes. Mol. Pharmacol. 3:142-152, 1967.

214a. Caggiano, V., R. Schnitzler, W. Strauss, R. K. Baker, A. C. Carter, A. S. Josephson, and S. Wallach. Zinc deficiency in a patient with retarded growth, hypogonadism, hypogammaglobulinemia and chronic infection. Amer. J. Med. Sci. 257:305-319, 1969.

215. Caines, L. A. The phosphorus content of some aquatic macrophytes with special reference to seasonal fluctuations and applications of phosphate fertilizers. Hydrobiologia 25:289-301, 1965.

216. Cairns, J., Jr., T. K. Bahns, D. T. Burton, K. L. Dickson, R. E. Sparks, and W. T. Waller. The effects of pH, solubility, and temperature upon the acute toxicity of zinc to the bluegill sunfish (Lepomis macrochirus Raf.). Trans. Kansas Acad. Sci. 74:81-92, 1972.

217. Cairns, J., Jr., and A. Scheier. The effects of periodic low oxygen upon the toxicity of various chemicals to aquatic organisms, pp. 165-176. In Proceedings of the Twelfth Industrial Waste Conference, 1957. Purdue University Engineering Extension Series No. 94. Lafayette, Ind.: Purdue University, 1958.

218. Calabrese, A., R. S. Collier, D. A. Nelson, and J. R. MacInnes. The toxicity of heavy metals to embryos of the American oyster Crassostrea virginica. Marine Biol. 18:162-166, 1973.

219. Caldwell, D. F., D. Oberleas, J. J. Clancy, and A. S. Prasad. Behavioral impairment in adult rats following acute zinc deficiency. Proc. Soc. Exp. Biol. Med. 133:1417-1421, 1970.

220. Caldwell, D. F., D. Oberleas, and A. S. Prasad. Reproductive performance of chronic mildly zinc deficient rats and the effects on behavior of their offspring. Nutr. Rep. Int. 7:309-319, 1973.

221. Calhoun, N. R., S. Campbell, Jr., and J. C. Smith, Jr. Accumulation of labeled zinc, strontium and calcium in bone injuries. J. Dent. Res. 49:1083-1085, 1970.

222. Callender, G. R., and C. J. Gentzkow. Acute poisoning by the zinc and antimony content of limeade prepared in a galvanized iron can. Milit. Surg. 80:67-71, 1937.

223. Calvery, H. O. Trace elements in food. Food Res. 7:313-331, 1942.

224. Calvin, H. I., C. C. Yu, and J. M. Bedford. Effects of epididymal maturation, zinc (II) and copper (II) on the reactive sulfhydryl content of structural elements in rat spermatozoa. Exp. Cell Res. 81:333-341, 1973.

225. Campbell, B. J., Y.-C. Lin, R. V. Davis, and E. Ballew. The purification and properties of a particulate renal dipeptidase. Biochim. Biophys. Acta 118:371-386, 1966.

226. Cann, J. R. Effect of ligands and oxidation state upon the reaction of myoglobin and hemoglobin with zinc. Biochemistry 3:903-908, 1964.

227. Cann, J. R. Kinetics in the reversible reaction of sperm whale myoglobin with zinc. Biochemistry 3:714-722, 1964.

228. Cannon, H. L. The use of plant indicators in ground water surveys, geologic mapping, and mineral prospecting. Taxon 20:227-256, 1971.

229. Cannon, H. L., and B. M. Anderson. The geochemists' involvement with the pollution problem, pp. 155-177. In H. L. Cannon and H. C. Hopps, Eds. Environmental Geochemistry in Health and Disease. The Geological Society of America, Inc., Memoir 123. Boulder, Colorado: The Geological Society of America, Inc., 1971.

230. Cares, J. W. The quantitative determination of airborne metallic dusts and fumes by x-ray spectrometry. Amer. Ind. Hyg. Assoc. J. 29:463-468, 1968.

231. Carey, A. G., Jr. Zinc-65 in benthic invertebrates off the Oregon coast, pp. 833-842. In A. T. Pruter and D. L. Alverson, Eds. The Columbia River Estuary and Adjacent Ocean Waters. Bioenvironmental Studies. Seattle: University of Washington Press, 1972.

232. Carleton, R. L., N. B. Friedman, and E. J. Bomze. Experimental teratomas of the testis. Cancer 6:464-473, 1953.

232a. Carlson, A. B. Contamination mechanisms, pp. 54-136. In J. A. Ayres, Ed. Decontamination of Nuclear Reactors and Equipment. New York: The Ronald Press, 1970.

233. Carolus, R. L. Distribution and redistribution of nutrients in perennial and biennial vegetables, pp. 202-209. In XVIth International Horticultural Congress -- 1962. Brussels, Belgium, Aug. 31-Sept. 8. Vol. 2. Gembloux, Belgium: J. Duculot, S. A., 1963.

234. Carpenter, F. H., and J. M. Vahl. Leucine aminopeptidase (bovine lens). Mechanism of activation by Mg^{2+} and Mn^{2+} of the zinc metalloenzyme, amino acid composition, and sulfhydryl content. J. Biol. Chem. 248:294-304, 1973.

235. Carpy, S. Inhibition de l'anhydrase carbonique erythrocytaire bovine B par differents agents chelateurs a pH 7.4. Biochim. Biophys. Acta 151:245-259, 1968.

236. Carrico, R. J., and H. F. Deutsch. The presence of zinc in human cytocuprein and some properties of the apoprotein. J. Biol. Chem. 245: 723-727, 1970.

237. Carroll, K. G., and J. L. Tullis. Observations on the presence of titanium and zinc in human leucocytes. Nature 217:1172-1173, 1968.

238. Carroll, M. D., and J. F. Loneragan. Response of plant species to concentrations of zinc in solution. I. Growth and zinc content of plants. Austral. J. Agric. Res. 19:859-868, 1968.

239. Carroll, M. D., and J. F. Loneragan. Response of plant species to concentrations of zinc in solution. II. Rates of zinc absorption and their relation to growth. Austral. J. Agric. Res. 20:457-463, 1969.

240. Carroll, R. E. The relationship of cadmium in the air to cardiovascular disease death rates. J.A.M.A. 198:267-269, 1966.

241. Carter, J. P., L. E. Grivetti, J. T. Davis, S. Nasiff, A. Mansour, W. A. Mousa, A.-E.-D. Atta, V. N. Patwardhan, M. A. Moneim, I. A. Abdou, and W. J. Darby. Growth and sexual development of adolescent Egyptian village boys. Effects of zinc, iron, and placebo supplementation. Amer. J. Clin. Nutr. 22:59-78, 1969.

242. Carter, M. J., and D. S. Parsons. The carbonic anhydrases of some guinea-pig tissues. Biochim. Biophys. Acta 206:190-192, 1970.

243. Carter, M. J., and D. S. Parsons. The purification and properties of carbonic anhydrases from guinea-pig erythrocytes and mucosae of the gastrointestinal tract. Biochem. J. 120:797-808, 1970.

244. Carvalho, A. P., and Y. Avivi. Effects of zinc on adenosine triphosphatase activity and superprecipitation of actomyosin from skeletal muscle of rabbit. Arch. Biochem. Biophys. 113:617-628, 1966.

245. Cash, W. D., and M. Gardy. Role of metal contaminants in the mitochondrial swelling activities of reduced and oxidized glutathione preparations. J. Biol. Chem. 240:3450-3452, 1965.

246. Catsch, A., and D. K. Lê. Removal of ^{60}Co and ^{65}Zn from the mammalian body. Experientia 21:724, 1965.

247. Catsch, A., and D. K. H. Lê. Das Verhalten von Radiozink-Chelaten im Säugetierorganismus. Strahlentherapie 130:557-566, 1966.

248. Caughey, J. E. Aetiological factors in adolescent malnutrition in Iran. N. Z. Med. J. 77:90-95, 1973.

249. Caughey, J. E. Zinc deficiency in man. Lancet 1:993, 1973. (letter)

250. Caughey, J. E. Zinc deficiency in man. Lancet 2:376-377, 1973. (letter)

251. Caujolle, F., Pham-Huu-Chanh, Nguyen-Luong-Thi-Hgo-Suong, and Phung van To. Recherches sur la toxicologie du zinc. 1. Toxicité immédiate et différée et toxicité à long terme. Agressologie 10:333-339, 1969.

252. Cawse, P. A., and D. H. Peirson. An Analytical Study of Trace Elements in the Atmospheric Environment. United Kingdom Atomic Energy Authority Research Group Report AERE-R7134. Harwell, Berkshire: Atomic Energy Research Establishment, Health Physics and Medical Division, 1972. 38 pp.

253. Cayla, J., and F. Fabre. La phosphatase sérique pendant la gestation. C. R. Soc. Biol. (Paris) 120:748-750, 1935.

254. Chan, W., and E. D. Brown. Plasma and bone zinc concentrations during zinc depletion. Fed. Proc. 34:906, 1975. (abstract)

255. Chandler, W. H., D. R. Hoagland, and P. L. Hibbard. Little-leaf or rosette in fruit trees. Proc. Amer. Soc. Hort. Sci. 28:556-560, 1931.

256. Chaney, R. L. Crop and food chain effects of toxic elements in sludges and effluents, pp. 129-141. In Proceedings of the Joint Conference on Recycling Municipal Sludges and Effluents on Land, July 9-13, 1973, University of Illinois, Champaign. Washington, D. C.: National Association of State Universities and Land-Grant Colleges, 1973.

257. Chang, T. L., T. A. Gover, and W. W. Harrison. Determination of magnesium and zinc in human brain tissue by atomic absorption spectroscopy. Anal. Chim. Acta 34:17-23, 1966.

257a. Chang, C. C., H. J. Tatum, and F. A. Kircl. The effect of intrauterine copper and other metals on implatation in rats and hamsters. Fertil. Steril. 21:274-278, 1970.

258. Chapman, H. D. Zinc, pp. 484-499. In H. D. Chapman, Ed. Diagnostic Criteria for Plants and Soils. Riverside: University of California, 1966.

259. Chapman, W. H., H. L. Fisher, and M. W. Pratt. Concentration Factors of Chemical Elements in Edible Aquatic Organisms. UCRL-50564. Livermore: University of California, Laurence Radiation Laboratory, 1968. 50 pp.

260. Chaube, S., H. Nishimura, and C. A. Swinyard. Zinc and cadmium in normal human embryos and fetuses. Arch. Environ. Health 26:237-240, 1973.

261. Chaudhry, F. M., and J. F. Loneragan. Zinc absorption by wheat seedlings. I. Inhibition by macronutrient ions in short-term experiments and its relevance to long-term zinc nutrition. Soil Sci. Soc. Amer. Proc. 36:323-327, 1972.

262. Chaudhry, F. M., and J. F. Loneragan. Zinc absorption by wheat seedlings. II. Inhibition by hydrogen ions and by micronutrient cations. Soil Sci. Soc. Amer. Proc. 36:327-331, 1972.

263. Chaudhry, F. M., M. Sharif, A. Latif, and R. H. Qureshi. Zinc-copper antagonism in the nutrition of rice (Oryza sativa L.). Plant Soil 38:573-580, 1973.

264. Li, C. The Chemical Arts of Old China. Easton, Penn.: Journal of Chemical Education, 1948. 215 pp.

265. Cheh, A. M., and J. B. Neilands. Role of zinc ion in beef liver ∫-aminolevulinate dehydratase. Fed. Proc. 33:1245, 1974. (abstract)

266. Chen, R. W., D. J. Eakin, and P. D. Whanger. Biological function of metallothionein. II. Its role in zinc metabolism in the rat. Nutr. Rep. Int. 10:195-200, 1974.

267. Chesnin, L. Corn, soybeans & other great plains crops, pp. [13-14]. In G. L. Berg, Ed. The Micronutrient Manual. Sponsored by Rayonier Incorporated, [1967].

267a. Chesters, J. K. Problems caused by variation in food intake in experiments on protein and nucleic acid metabolism. Proc. Nutr. Soc. 30: 1-6, 1971.

268. Chesters, J. K. Biochemical functions of zinc with emphasis on nucleic acid metabolism and cell division, pp. 39-50. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium on Trace Element Metabolism in Animals held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

269. Chesters, J. K. The role of zinc ions in the transformation of lymphocytes by phytohaemagglutinin. Biochem. J. 130:133-139, 1972.

270. Chesters, J. K., and J. Quarterman. Effects of zinc deficiency on food intake and feeding patterns of rats. Brit. J. Nutr. 24:1061-1069, 1970

271. Chesters, J. K., and M. Will. Some factors controlling food intake by zinc-deficient rats. Brit. J. Nutr. 30:555-566, 1973.

272. Chew, R. M., and J. G. Hemington. Turnover of zinc-65 as an index of energy metabolism of Perognathus longimembris, the little pocket mouse, pp. 247-252. In D. J. Nelson, Ed. Radionuclides in Ecosystems. Vol. 1. Proceedings of the Third National Symposium on Radioecology, May 10-12, 1971, Oak Ridge, Tennessee. CONF-710501. Oak Ridge: U. S. Atomic Energy Commission, (not dated).

273. Chiaudani, G. Contenuti normali ed accumuli di rame in Phragmites communis L. come risposta a quelli nei sedmenti di sei laghi Italiani. Mem. Istit. Ital. Idrobiol. 25:81-95, 1969. (summary in English)

274. Chipperfield, B., and J. R. Chipperfield. Heart-muscle magnesium, potassium, and zinc concentrations after sudden death from heart-disease. Lancet 2:293-296, 1973.

74a. Chipperfield, B., J. R. Chipperfield, G. Behr, and P. Burton. Magnesium and potassium content of normal heart muscle in areas of hard and soft water. Lancet 1:121-122, 1976.

275. Chipman, W. A., T. R. Rice, and T. J. Price. Uptake and accumulation of radioactive zinc by marine plankton, fish and shellfish. U. S. Fish Wildlife Serv. Fish. Bull. 58:279-292, 1958.

275a. Chlebowski, J. F., and J. E. Coleman. Zinc and its role in enzymes, pp. 2-140. In H. Sigel, Ed. Metal Ions in Biological Systems. Vol. 6. Biological Action of Metal Ions. New York: Marcel Dekker, Inc., 1976.

276. Christensen, C. M., D. F. Caldwell, and D. Oberleas. Establishment of a learned preference for a zinc-containing solution by zinc-deficient rats. J. Comp. Physiol. Psychol. 87:415-421, 1974.

277. Christensen, R. E., R. M. Beckman, and J. J. Birdsall. Spices and other condiments. Some mineral elements of commercial spices and herbs as determined by direct reading emission spectroscopy. J. Assoc. Off. Anal. Chem. 51:1003-1010, 1968.

278. Christian, G. D., and E. J. Feldman. Copper, silver, gold, pp. 331-339. In Atomic Absorption Spectroscopy. Applications in Agriculture, Biology and Medicine. New York: Wiley-Interscience, 1970.

279. Christian, G. D., and F. J. Feldman. Zinc, cadmium, mercury, pp. 347-364. In Atomic Absorption Spectroscopy. Applications in Agriculture, Biology and Medicine. New York: Wiley-Interscience, 1970.

280. Chu, R. C., and D. H. Cox. Excessive dietary zinc during lactation and nutritive value and mineral composition of rat's milk. Nutr. Rep. Int. 2:179-184, 1970.

281. Chu, R. C., and D. H. Cox. Zinc, iron, copper, calcium, cytochrome oxidase, and phospholipid in rats of lactating mothers fed excess zinc. Nutr. Rep. Int. 5:61-66, 1972.

282. Chu, R. C., S. A. Schlicker, and D. H. Cox. A zinc-biotin interrelationship in the rat. Nutr. Rep. Int. 1:11-18, 1970.

283. Chumbley, C. G. Permissible Levels of Toxic Metals in Sewage Used on Agricultural Land. A.D.A.S. (Agricultural Development and Advisory Service) Advisory Paper No. 10. Middlesex: Great Britain Ministry of Agriculture, Fisheries and Food, 1971. 12 pp.

284. Chunn, V. D., Jr. Metal toys containing zinc, and anemia in children. Ped. Dig. 15:20-24, 1973.

285. Chvapil, M. New aspects in the biological role of zinc: A stabilizer of macromolecules and biological membranes. Life Sci. 13:1041-1049, 1973.

286. Chvapil, M., J. N. Ryan, and C. F. Zukoski. The effect of zinc and other metals on the stability of lysosomes. Proc. Soc. Exp. Biol. Med. 140:642-646, 1972.

287. Clark, B., and J. W. Porteous. The metal ion activation of the alkaline β-glycerophosphatase of rabbit small intestine. Biochem. J. 95: 475-482, 1965.

288. Clark, J. L., W. H. Pfander, and G. B. Thompson. Urea and trace minerals for finishing cattle rations. J. Anim. Sci. 30:297-302, 1970.

289. Clark, M. C., and D. J. Swaine. Trace Elements in Coal. I. New South Wales Coal. II. Origin, Mode of Occurrence, and Economic Importance. Commonwealth of Australia. Commonwealth Scientific and Industrial Research Organization. Division of Coal Research. Technical Communication 45, 1962. 112 pp.

290. Clarke, A. L., and E. R. Graham. Zinc diffusion and distribution coefficients in soil as affected by soil texture, zinc concentration and pH. Soil Sci. 105:409-418, 1968.

291. Clarke, E. G. C. Poisoning in the pig. Brit. Vet. J. 125:289-293, 1969.

291a. Clark, L. J., and W. L. Hill. Occurrence of manganese, copper, zinc, molybdenum, and cobalt in phosphate fertilizers and sewage sludge. J. Assoc. Off. Agric. Chem. 41:631-637, 1958.

291b. Clayton, R. J., Double-blind trial of oral zinc sulphate in patients with leg ulcers. Brit. J. Clin. Pract. 26:368-370, 1972.

292. Cocucci, M. C., and G. Rossi. Biochemical and morphological aspects of zinc deficiency in Rhodotorula gracilis. Arch. Mikrobiol. 85:267-279, 1972.

293. Deleted

294. Cohanim, M., and E. R. Yendt. Zinc excretion in patients with renal calculi. Clin. Res. 19:807, 1971. (abstract)

295. Cohen, C. Zinc sulphate and bedsores. Brit. Med. J. 2:561, 1968. (letter)

296. Cohen, I. K., P. J. Schechter, and R. I. Henkin. Hypogeusia, anorexia and altered zinc metabolism following thermal burn. J.A.M.A. 223: 914-916, 1973.

297. Cohen, J. M., L. J. Kamphake, E. K. Harris, and R. L. Woodward. Taste threshold concentrations of metals in drinking water. J. Amer. Water Works Assoc. 52:660-670, 1960.

298. Colburn, R. W., and J. W. Maas. Adenosine triphosphate-metal-norepinephrine ternary complexes and catecholamine binding. Nature 208:37-41, 1965.

299. Coleman, C. B., and G. Matrone. In vivo effect of zinc on iron-induced ferritin synthesis in rat liver. Biochim. Biophys. Acta 177:106-112, 1969.

300. Coleman, B. W., E. M. Reimann, R. H. Grummer, M. L. Sunde, and W. G. Hoekstra. Antagonistic effect of arginine on zinc metabolism in chicks. J. Nutr. 101:1695-1702, 1971.

301. Coleman, J. E. Human carbonic anhydrase. Protein conformation and metal ion binding. Biochemistry 4:2644-2655, 1965.

302. Coleman, J. E. Metal ion dependent binding of sulphonamide to carbonic anhydrase. Nature 214:193-194, 1967.

303. Coleman, J. E. Metal ions in enzymatic catalysis. Prog. Bioorganic Chem. 1:159-344, 1971.

304. Coleman, J. E. The reactivities of functional groups of metalloproteins. MTP Int. Rev. Sci. Biochem. Ser. 1:185-260, 1974.

305. Coleman, J. E., P. Pulido, and B. L. Vallee. Organic modifications of metallocarboxypeptidases. Biochemistry 5:2019-2026, 1966.

306. Coleman, J. E., and B. L. Vallee. Metallocarboxypeptidase-inhibitor complexes. Biochemistry 3:1874-1879, 1964.

307. Colovos, G., G. S. Wilson, and J. Moyers. Determination of trace amounts of zinc, cadmium, lead and copper in airborne particulate matter by anodic stripping voltammetry. Anal. Chim. Acta 64:457-464, 1973.

307a. Comens, P. Experimental hydralazine disease and its similarity to disseminated lupus erythematosus. J. Lab. Clin. Med. 47:444-454, 1956.

307b. Comens, P. Manganese depletion as an etiological factor in hydralazine disease. Amer. J. Med. 20:944-945, 1956. (abstract)

307c. Comens, P. Chronic intoxication from hydralazine resembling disseminated lupus erythematosus and its apparent reversal by manganese, pp. 312-320. In M. J. Seven and L. A. Johnson, Eds. Metal-Binding in Medicine. Proceedings of a Symposium. Philadelphia: J. B. Lippincott Company, 1960.

308. Condon, C. J., and R. M. Freeman. Zinc metabolism in renal failure. Ann. Intern. Med. 73:531-536, 1970.

309. Connor, P. M. Acute toxicity of heavy metals to some marine larvae. Marine Pollut. Bull. 3:190-192, 1972.

309a. Consolazio, C. F., R. A. Nelson, L. R. Matoush, R. C. Hughes, and P. Urone. The Trace Mineral Losses in Sweat. Report No. 284, U. S. Army Medical Research and Nutrition Laboratory, Denver, Colorado, 1964. 14 pp.

310. Constam, G. R., W. Leemann, F. Almasy, and A. G. Constam. Weitere Beobachtungen über den Zinkstoffwechsel bei Diabetes mellitus. Schweiz. Med. Wochenschr. 94:1104-1109, 1964.

311. Conte, N., and C. Scandellari. Studies on zinc metabolic. II. Kinetics of Zn^{65} in the organs of normal rabbits. Acta Isotop. 3:367-375, 1963.

312. Coombs, T. L. The distribution of zinc in the oyster Ostrea edulis and its relation to enzymic activity and to other metals. Marine Biol. 12: 170-178, 1972.

313. Coombs, T. L., Y. Omote, and B. L. Vallee. The zinc binding groups of carboxypeptidase A. Biochemistry 3:653-662, 1964.

314. Cooper, J. A. Comparison of particle and photon excited x-ray fluorescence applied to trace element measurements on environmental samples. Nucl. Instrum. Meth. 106:525-538, 1973.

315. Cooper, S. S., and M. L. Sullivan. Spectrophometric studies of dithizone and some dithizonates. Anal. Chem. 23:613-618, 1951.

316. Corder, C. N., and O. H. Lowry. The zinc requirement of dinucleotide pyrophosphatase in mammalian organs. Biochim. Biophys. Acta 191: 579-587, 1969.

317. Corn, M. Nonviable particles in air, pp. 47-94. In A. C. Stern, Ed. Air Pollution. Vol. 1. Air Pollution and Its Effects. (2nd ed.) New York: Academic Press, 1968.

318. Cornec, E. Étude spectrographique des cendres de plantes marines. C. R. Acad. Sci. (Paris) 168:513-514, 1919.

319. Cosgrove, J. F., and D. J. Braco. Determination of minor metallic elements in the water environment, pp. 1315-1356. In L. L. Ciaccio, Ed. Water and Water Pollution Handbook. Vol. 4. New York: Marcel Dekker, Inc., 1973.

320. Cotterill, C. H. Distilling, pp. 52-57. In Industrial Plant Location. Its Application to Zinc Smelting. Saint Louis: American Zinc, Lead & Smelting Co., 1950.

321. Cotterill, C. H., and J. M. Cigan, Eds. Extractive Metallurgy of Lead and Zinc. Vol. 2 of AIME World Symposium on Mining & Metallurgy of Lead & Zinc. New York: American Institute of Mining, Metallurgical and Petroleum Engineers, Inc., 1970. 1090 pp.

322. Cotzias, G. C., D. C. Borg, and B. Selleck. Specificity of zinc pathway in the rabbit: Zinc-cadmium exchange. Amer. J. Physiol. 201:63-66, 1961.

323. Cotzias, G. C., and P. S. Papavasiliou. Specificity of zinc pathway through the body: Homeostatic considerations. Amer. J. Physiol. 206:787-792, 1964.

324. Cousins, R. J., A. K. Barber, and J. R. Trout. Cadmium toxicity in growing swine. J. Nutr. 103:964-972, 1973.

325. Cowling, H., and E. J. Miller. Determination of small zmounts of zinc in plant materials. A photometric dithizone method. Ind. Eng. Chem. Anal. Ed. 13:145-149, 1941.

326. Cox, D. H. Excess dietary zinc and subcellular changes in hepatic zinc, iron, and copper in maternal and fetal rats. Nutr. Rep. Int. 5:145-150, 1972.

327. Cox, D. H., R. C. Chu, and S. A. Schlicker. Zinc deficiency in the maternal rat during gestation, and zinc, iron, copper, and calcium content and enzyme activity in maternal and fetal tissues. J. Nutr. 98:449-458, 1969.

328. Cox, D. H., and O. M. Hale. Liver iron depletion without copper loss in swine fed excess zinc. J. Nutr. 77:225-228, 1962.

329. Cox, D. H., and D. L. Harris. Effect of excess dietary zinc on iron and copper in the rat. J. Nutr. 70:514-520, 1960.

330. Cox, D. H., S. A. Schlicker, and R. C. Chu. Excess dietary zinc for the maternal rat, and zinc, iron, copper, calcium, and magnesium content and enzyme activity in maternal and fetal tissues. J. Nutr. 98:459-466, 1969.

331. Cox, R. P. Hormonal stimulation of zinc uptake in mammalian cell cultures. Mol. Pharmacol. 4:510-521, 1968.

332. Craft, L. R. Isolation of a zinc protein from the tapetum lucidum of the cat. Biochem. J. 130:303-305, 1972.

332a. Craun, G. F., and L. J. McCabe. Problems associated with metals in drinking water. J. Amer. Water Works Assoc. 67:593-599, 1975.

333. Cragle, R. G. Dynamics of mineral elements in the digestive tract of ruminants. Fed. Proc. 32:1910-1014, 1973.

333a. Crawford, I. L., and J. D. Connor. Zinc in maturing rat brain: Hippocampal concentration and localization. J. Neurochem. 19:1451-1458, 1972.

333b. Crawford, T., and M. D. Crawford. Prevalence and pathological changes of ischaemic heart-disease in a hard-water and in a soft-water area. Lancet 1:229-232, 1967.

334. Crawford, M. D. Hardness of drinking-water and cardiovascular disease. Proc. Nutr. Soc. 31:347-353, 1972.

335. Creason, J. P., O. McNulty, L. T. Heiderscheit, D. H. Swanson, and R. W. Beuchley. Roadside gradients in atmospheric concentrations of cadmium, lead, and zinc, pp. 129-142. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - V. Proceedings of 5th Annual Conference on Trace Substance in Environmental Health held in Columbia, Missouri, June 29 - July 1, 1971. Columbia: University of Missouri, 1972.

336. Cristol, M. P. Le zinc dans les tissus cancéreux. Contribution à l'étude de la physiopathologie du zinc, et, en particulier, de sa signification dans les tumeurs. Bull. Soc. Chim. Biol. 5:23-40, 1923.

337. Cross, F. A., and J. H. Brooks. Concentrations of manganese, iron, and zinc in juveniles of five estuarine-dependent fishes, pp. 769-775. In D. J. Nelson, Ed. Radionuclides in Ecosystems. Vol. 2. Proceedings of the Third National Symposium on Radioecology, May 10-12, 1971, Oak Ridge, Tennessee. CONF-710501. Oak Ridge: U. S. Atomic Energy Commission, (not dated).

338. Cross, F. A., J. M. Dean, and C. L. Osterberg. The effect of temperature, sediment and feeding on the behavior of four radionuclides in a marine benthic amphipod, pp. 450-461. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

339. Cross, F. A., T. W. Duke, and J. N. Willis. Biogeochemistry of trace elements in a coastal plain estuary: Distribution of manganese, iron and zinc in sediments, water and polychaetous worms. Chesapeake Sci. 11:221-234, 1970.

340. Cross, F. A., S. W. Fowler, J. M. Dean, L. F. Small, and C. L. Osterberg. Distribution of ^{65}Zn in tissues of two marine crustaceans determined by autoradiography. J. Fish. Res. Board Can. 25:2461-2466, 1968.

341. Cross, F. A., L. H. Hardy, N. Y. Jones, and R. T. Barber. Relation between total body weight and concentrations of manganese, iron, copper, zinc and mercury in white muscle of bluefish (Pomatomus saltatrix) and a bathyl-demersal fish Antimora rostrata. J. Fish. Res. Board Can. 30:1287-1291, 1973.

342. Cross, F. A., J. N. Willis, L. H. Hardy, N. Y. Jones, and J. M. Lewis. Role of juvenile fish in cycling of Mn, Fe, Cu, and Zn in a coastal-plain estuary, pp. 45-63. In Mineral Cycling in Southeastern Ecosystems. Proceedings of a Symposium, 1974. CONF-740513. Oak Ridge, Tenn.: U. S. Energy Research and Development Administation, 1975.

343. Cohanim, M., and E. R. Yendt. The effects of thiazides on serum and urinary zinc in patients with renal calculi. Johns Hopkins Med. J. 136:137-141, 1975.

344. Cumings, J. N. Trace metals of the brain and their importance in human disease. Chem. Weekblad 63:473-479, 1967.

344a. Cushing, C. E. Concentration and transport of ^{32}P and ^{65}Zn by Columbia River plankton. Limnol. Oceanogr. 12:330-332, 1967.

344b. Cunliffe, W. J., and S. Shuster. The rate of sebum excretion in man. Brit. J. Derm. 81:697-704, 1969.

345. Cushing, C. E., and F. L. Rose. Cycling of zinc-65 by Columbia River periphyton in a closed microcosm. Limnol. Oceanogr. 15:762-767, 1970.

346. Cushing, C. E., and D. G. Watson. Accumulation of ^{32}P and ^{65}Zn by living and killed plankton. Oikos 19:143-145, 1968.

346a. Cusworth, D. C., C. E. Dent, and C. R. Scriver. Primary hyperparathyroidism and hyperaminoaciduria. Clin. Chim. Acta 41:355-361, 1972.

347. Cutshall, N. Turnover of zinc-65 in oysters. Health Phys. 26:327-331, 1974.

348. Czerniejewski, C. P., C. W. Shank, W. G. Bechtel, and W. B. Bradley. The minerals of wheat, flour, and bread. Cereal Chem. 41:65-72, 1964.

349. Dahmer, E. J., B. W. Coleman, R. H. Grummer, and W. G. Hoekstra. Alleviation of parakeratosis in zinc deficient swine by high levels of dietary histidine. J. Anim. Sci. 35:1181-1189, 1972.

350. Dahmer, E. J., R. H. Grummer, and W. G. Hoekstra. Prevention of zinc deficiency in swine by feeding blood meal. J. Anim. Sci. 35:1176-1180, 1972.

351. Dale, I. M., H. J. Duncan, and C. McDonald. Neutron activation analysis of atmospheric particulates. Radiochem. Radioanal. Lett. 15:77-86, 1973.

351a. Dallman, P. R., R. A. Spirito, and M. A. Siimes. Diurnal patterns of DNA synthesis in the rat: Modification by diet and feeding schedule. J. Nutr. 104:1234-1241, 1974.

352. Dams, R., K. A. Rahn, J. A. Robbins, G. D. Nifong, and J. W. Winchester. Multi-element analysis of air pollution particulates by nondestructive neutron activation, pp. 509-516. In H. J. Englund and W. T. Beery, Eds. Proceedings of the Second International Clean Air Congress. Held at Washington, D.C., Dec. 6-11, 1970. New York: Academic Press, 1971.

353. Dams, R., K. A. Rahn, and J. W. Winchester. Evaluation of filter materials and impaction surfaces for nondestructive neutron activation analysis of aerosols. Environ. Sci. Technol. 6:441-448, 1972.

353a. Damyanov, I., and W. Dutz. Anencephaly in Shriaz, Iran. Lancet 1:82, 1971. (letter)

354. Dams, R., J. A. Robbins, K. A. Rahn, and J. W. Winchester. Nondestructive neutron activation analysis of air pollution particulates. Anal. Chem. 42:861-867, 1970.

354a. Daniel, E. E., S. Fair, and A. M. Kidwai. Zinc and smooth muscle contractility. II. Zinc content and accumulation in rat uterus and their relation to contractility changes from zinc and ouabain. J. Pharmacol. Exp. Ther. 178:290-298, 1971.

354b. Daniel, E. E., S. Fair, A. M. Kidwai, and I. Polacek. Zinc and smooth muscle contractility. I. Study of the mechanisms of zinc-induced contractility changes in rat uteri. J. Pharmacol. Exp. Ther. 178: 282-289, 1971.

354c. Danbolt, N., and K. Closs. Akrodermatitis enteropathica. Acta Derm. Venereol. 23:127-169, 1942-43.

354d. Danielsen, A., and E. Steinnes. A study of some selected trace elements in normal and cancerous tissue by neutron activation analysis. J. Nucl. Med. 11:260-264, 1970.

355. Daniel, O., F. Haddad, G. Prout, and W. F. Whitmore. Some observations on the distribution of radioactive zinc in prostatic and other human tissues. Brit. J. Urol. 28:271-278, 1956.

356. Dart, E. E. [Reaction of workers exposed to zinc oxide fumes], p. 517. In F. A. Patty, Ed. Industrial Hygiene and Toxicology. Vol. 1. New York: Interscience Publishers, Inc., 1948.

357. David, J. J., and R. F. Foster. Bioaccumulation of radioisotopes through aquatic food chains. Ecology 39:530-535, 1958.

358. Davies, A. G. The kinetics of and a preliminary model for the uptake of radio-zinc by Phaeodactylum tricornutum in culture, pp. 403-420. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA, in Seattle, U.S.A., 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

359. Davies, B. E. Trace metal content of soils affected by base metal mining in the west of England. Oikos 22:366-372, 1971.

360. Davies, I. J. T. Plasma-zinc concentrations in patients with bronchogenic carcinoma. Lancet 1:149, 1972. (letter)

361. Davies, I. J. T., M. Musa, and T. L. Dormandy. Measurements of plasma zinc. Part I. In health and disease. J. Clin. Path. 21:359-365, 1968.

361a. Davies, M. I., and I. Motzok. Intestinal alkaline phosphatase and phytase of chicks: Separation of isoenzymes, zinc contents and in vitro effects of zinc. Comp. Biochem. Physiol. 42B:345-356, 1972.

361b. Davies, M. I., and I. Motzok. Zinc deficiency in the chick: Effect on tissue alkaline phosphatases. Comp. Biochem. Physiol. 40B:129-137, 1971.

362. Davies, N. T. Intestinal transport of zinc by rat duodenum. J. Physiol. 229:46P-47P, 1972.

362a. Davies, N. T. Recent studies of antagonistic interactions in the actiology of trace element dificiency and excess. Proc. Nutr. Soc. 33:293-298, 1974.

362b. Davies, N. T., and R. Nightingale. The effect of phytate on intestinal absorption and secretion of zinc, and whole-body retention of zinc, copper, iron and manganese in rats. Brit. J. Nutr. 34:243-258, 1975.

362c. Davies, I. J. T. The Clinical Significance of the Essential Biological Metals. London: William Heinemann Medical Books, Ltd., 1972. 126 pp.

363. Davies, N. T. The effect of protein synthesis inhibitors on zinc absorption by rat duodenum in vivo. J. Physiol. 244:38P-39P, 1975.

364. Davies, N. T., I. Bremner, and C. F. Mills. Studies on the induction of a low-molecular-weight zinc-binding protein in rat liver. Biochem. Soc. Trans. 1:985-988, 1973.

365. Davis, G. K. Competition among mineral elements relating to absorption by animals. Ann. N. Y. Acad. Sci. 199:62-69, 1972.

366. Davis, J. J., and F. R. Foster. Bioaccumulation of radioisotopes through aquatic food chains. Ecology 39:530-535, 1958.

367. Davis, P. N., C. C. Norris, and F. H. Kratzer. Interference of soybean proteins with the utilization of trace metals. J. Nutr. 77:217-223, 1962.

368. Davis, W. E., and Associates. Consumptive uses of rubber tires and motor oil, pp. 36-37. In National Inventory of Sources and Emissions: Cadmium, Nickel and Asbestos--1968. Cadmium, Section I. Leawood, Kans.: W. E. Davis and Associates, Feb. 1970.

369. Davis, W. E., and Associates. National Inventory of Sources and Emissions. Zinc. APTD-1139. Leawood, Kansas: W. E. Davis and Associates, 1972. 77 pp.

370. Dawkins, J. M. Zinc and Spelter. Oxford: Zinc Development Association, 1950. p. 5.

371. Day, R., and J. Franklin. Plant carbonic anhydrase. Science 104:363-365, 1946.

371a. Dean, R. B., and J. E. Smith, Jr. The properties of sludges, pp. 39-47. In Proceedings of the Joint Conference on Recycling Municipal Sludges and Effluents on Land. Champaign, Illinois, July 9-13, 1973. Washington, D. C.: National Association of State Universities and Land Grant Colleges, 1973.

372. DeKock, P. C., and R. L. Mitchell. Uptake of chelated metals by plants. Soil Sci. 84:55-62, 1957.

373. Delves, H. T. A micro-sampling method for the rapid determination of lead in blood by atomic-absorption spectrophotometry. Analyst 95: 431-438, 1970.

373a. Delves, H. T., F. W. Alexander, and H. Lay. Copper and zinc concentration in the plasma of leukaemic children. Brit. J. Haematol. 24:525-531, 1973.

374. Demertzis, P. N. Effect of zinc on skin and hair. Lancet 2:1261-1262, 1972. (letter)

375. Demertzis, P. N., and C. F. Mills. Oral zinc therapy in the control of infectious pododermatitis in young bulls. Vet. Rec. 93:219-222, 1973.

376. Dennes, E., R. Tupper, and A. Wormall. Studies on zinc in blood. Transport of zinc and incorporation of zinc in leucocytes. Biochem. J. 82:466-476, 1962.

377. Dennes, E., R. Tupper, and A. Wormall. The zinc content of erythrocytes and leucocytes of blood from normal and leukaemic subjects. Biochem. J. 78:578-587, 1961.

378. DeRemer, E. D., and R. L. Smith. A preliminary study on the nature of a zinc deficiency in field beans as determined by radioactive zinc. Agron. J. 56:67-70, 1964.

378a. Derrien, E., and C. Benoît. Notes et observations sur les urines et sur quelques organes d'un femme morte en crise de porphyrie aiguë. Arch. Soc. Sci. Med. Biol. (Montpellier) 10:456-472, 1929.

379. DeVoe, J. R., Ed. Modern Trends in Activation Analysis. Proceedings of the 1968 International Conference held at the National Bureau of Standards, Gaithersburg, Maryland, Oct. 7-11, 1968. NBS Special Publication 312. Vols. I & II. Washington, D. C.: U. S. Government Printing Office, 1969. 1334 pp.

379a. Deyl, Z., J. Rosmus, and M. Adam. Investigation on the reaction of metals with collagen in vivo. Eur. J. Biochem. 13:589-592, 1970.

380. DeWys, W., and W. Pories. Inhibition of a spectrum of animal tumors by dietary zinc deficiency. J. Nat. Cancer Inst. 48:375-381, 1972.

381. DeWys, W., W. J. Pories, M. C. Richter, and W. H. Strain. Inhibition of Walker 256 carcinosarcoma growth by dietary zinc deficiency. Proc. Soc. Exp. Biol. Med. 135:17-22, 1970.

382. Diamond, I., and L. S. Hurley. Histopathology of zinc-deficient fetal rats. J. Nutr. 100:325-329, 1970.

382a. Delete 382a--use 383.

383. Diamond, I., H. Swenerton, and L. S. Hurley. Testicular and esophageal lesions in zinc-deficient rats and their reversibility. J. Nutr. 101:77-84, 1971.

383a. Dieleman, L. S., and B. A. Underwood. The role of zinc in the mobilization of vitamin A in the rat. Fed. Proc. 34:919, 1975. (abstract)

384. Dietz, F. The enrichment of heavy metals in submerged plants, pp. 53-62. In S. H. Jenkins, Eds. Advances in Water Pollution Research. Proceedings of the Sixth International Conference held in Jerusalem, June 18-23, 1972. New York: Pergamon Press, 1973.

384a. Dillaha, C. J., A. L. Lorincz, and O. R. Aavik. Acrodermatitis enteropathica. Review of the literature and report of a case successfully treated with diodoquin. J.A.M.A. 152:509-512, 1953.

385. Dines, D. E., L. R. Elveback, and J. T. McCall. Zinc, copper and iron content of pleural fluid in benign and neoplastic disperse. Thorax 27:368-370, 1972.

386. Dittrich, T. R., and C. R. Cothern. Analysis of trace metal particulates in atmospheric samples using x-ray fluorescence. J. Air Pollut. Control Assoc. 21:716-719, 1971.

387. Dixit, P. K., and A. Lazarow. Effects of metal ions and sulfhydryl inhibitors on glucose metabolism by adipose tissue. Amer. J. Physiol. 213:849-856, 1967.

388. Dobry-Duclaux, A. Structure de centre active de l'anhydrase carbonique. C. R. Acad. Sci. D (Paris) 261:2131-2133, 1965.

389. Dobry-Duclaux, A. Sur la détermination des sites actifs de certains enzymes. VIII. -- Sur les chélates-modèles du centre actif de l'anhydrase carbonique. Bull. Soc. Chim. Biol. 48:895-903, 1966.

390. Doby-Duclaux, A. Sur la détermination des sites de certains enzymes. VI. Étude de l'inhibition dans les deux sens de la réaction. Biochim. Biophys. Acta 89:1-22, 1964.

391. Dodson, E., M. M. Harding, D. C. Hodgkin, and M. G. Rossmann. The crystal structure of insulin. III. Evidence for a 2-fold axis in rhombohedral zinc insulin. J. Mol. Biol. 16:227-241, 1966.

392. Dolar, S. G., D. R. Keeney, and G. Chesters. Mercury accumulation by Myriophyllum spicatum L. Environ. Lett. 1:191-198, 1971.

393. Donaldson, J., T. St. Pierre, J. L. Minnich, and A. Barbeau. Determination of Na^+, K^+, Mg^{2+}, Cn^{2+}, Zn^{2+}, and Mn^{2+} in rat brain regions. Can. J. Biochem. 51:87-92, 1973.

393a. Dorn, C. R., P. E. Phillips, J. O. Pierce, II, and G. R. Chase. Cadmium, copper, lead, and zinc in bovine hair in the new lead belt of Missouri. Bull. Environ. Contam. Toxicol. 12:626-632, 1974.

394. Doudoroff, P., and M. Katz. Critical review of literature on the toxicity of industrial wastes and their components to fish. II. The metals, as salts. Sewage Ind. Wastes 25:802-839, 1953.

395. Dowdy, R. P., and F. H. Nielsen. Effect of histidine, histamine, and aspirin on sulfur-35 metabolism in zinc-deficient chick bone. J. Nutr. 102:529-534, 1972.

396. Drake, M. Soil chemistry and plant nutrition, pp. 395-444. In F. E. Bear, Ed. Chemistry of the Soil. (2nd ed.) American Chemical Society Monograph Series 160. New York: Reinhold Publishing Corporation, 1964.

397. Dreosti, I. E., P. C. Grey, and P. J. Wilkins. Deoxyribonucleic acid synthesis, protein synthesis and teratogenesis in zinc-deficient rats. S. Afr. Med. J. 46:1585-1588, 1972.

398. Drinker, C. K., and L. T. Fairhill. Zinc in relation to general and industrial hygiene. Public Health Rep. 48:955-961, 1933.

399. Drinker, K. R., and P. Drinker. Metal fume fever: V. Results of the inhalation by animals of zinc and magnesium oxide fumes. J. Ind. Hyg. 10:56-70, 1928.

400. Drinker, K. R., P. K. Thompson, and M. Marsh. An investigation of the effect upon rats of long-continued ingestion of zinc compounds, with especial reference to the relation of zinc excretion to zinc intake. Amer. J. Physiol. 81:284-306, 1927.

401. Drinker, K. R., P. K. Thompson, and M. Marsh. An investigation of the effect of long-continued ingestion of zinc, in the form of zinc oxide, by cats and dogs, together with observations upon the excretion and the storage of zinc. Amer. J. Physiol. 80:31-64, 1927.

402. Drinker, P. Certain aspects of the problem of zinc toxicity. J. Ind. Hyg. 4:177-197, 1922.

402a. Drinker, P., R. M. Thomson, and R. L. Finn. Metal fume fever: IV. Threshold doses of zinc oxide, preventive measures, and the chronic effects of repeated exposures. J. Ind. Hyg. 9:331-345, 1927.

403. Drinnan, R. E. Observations on the Accumulation of Heavy Metals by Shellfish in the Estuary of the Miramichi River, N.B. Manuscript Report Series (Biological), Ellerslee Sub-Station, Biological Station, St. Andrews, N. B., Fisheries Research Board of Canada, 1966. 15 pp.

404. Droste, H., and F. Jekat. Nährstoffversorgung in Arbeitnehmer- und Rentnerhaushalten der Bundesrepublik Deutschland während der Zeit 1950-1962. II. Die Zufuhr von Mineralien und Ballaststoffen, Vitaminen und Aminosäuren. Nutr. Diet. 7:1-20, 1965.

405. Duff, T. A., and J. E. Coleman. Macaca mulata carbonic anhydrase. Crystallization and physicochemical and enzymatic properties of two isozymes. Biochemistry 5:2009-2019, 1966.

406. Duke, T., J. Willis, T. Price, and K. Fischler. Influence of environmental factors on the concentration of ^{65}Zn by an experimental community, pp. 355-362. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

407. Duncan, G. D., L. F. Gray, and L. J. Daniel. Effect of zinc on cytochrome oxidase activity. Proc. Soc. Exp. Biol. Med. 83:625-627, 1953.

408. Duncan, J. R., and I. E. Dreosti. The effect of zinc deficiency on the timing of deoxyribonucleic acid synthesis in regenerating rat liver. S. Afr. Med. J. 48:1697-1699, 1974.

409. Durum, W. H., J. D. Hem, and S. G. Heidel. Reconnaissance of Selected Minor Elements in Surface Waters of the United States, October 1970. U. S. Geological Survey Circular 643. Washington, D. C.: U. S. Department of the Interior, 1971. 49 pp.

410. Dvorak, H. F., and L. A. Heppel. Metallo-enzymes released from Escherichia coli by osmotic shock. II. Evidence that 5'- nucleotidase and cyclic phosphodiesterase are zinc metalloenzymes. J. Biol. Chem. 243:2647-2653, 1968.

411. Eads, E. A., and C. E. Lambdin. A survey of trace metals in human hair. Environ. Res. 6:247-252, 1973.

412. Earle, I. P., and J. W. Stevenson. Relation of dietary zinc to composition of sow colostrum and milk. J. Anim. Sci. 24:325-328, 1965.

413. Earley, E. B. Minor element studies with soybeans: I. Varietal reaction to concentrations of zinc in excess of nutritional requirement. J. Amer. Soc. Agron. 35:1012-1023, 1943.

414. Eckert, H. Über die Austauschbarkeit des Zinks gegen andere Schwermetalle in der Molekel der Kohlensäureanhydratase. (KA) Naturwissenschaften 48:429-430, 1961.

414a. Eddington, C. L., H. F. Upchurch, and R. F. Kampschmidt. Effect of extracts from rabbit leukocytes on levels of acute phase globulins in rat serum. Proc. Soc. Exp. Biol. Med. 136:159-164, 1970.

415. Edwards, G. E., and A. K. Mohamed. Reduction in carbonic anhydrase activity in zinc deficient leaves of Phaseolus vulgaris L. Crop Sci. 13:351-354, 1973.

416. Edwards, H. M., Jr. The availability to chicks of zinc in various compounds and ores. J. Nutr. 69:306-308, 1959.

417. Edwards, R. R. C. Estimation of the respiratory rate of young plaice (Pleuronectes platessa L.) in natural conditions using zinc-65. Nature 216:1335-1337, 1967.

418. Egan, A. R. Reproductive responses to supplemental zinc and manganese in grazing Dorset Horn ewes. Austral. J. Exp. Agric. Anim. Husb. 12: 131-135, 1972.

419. Eggleton, W. G. E. The zinc and copper contents of the organs and tissues of Chinese subjects. Biochem. J. 34:991-997, 1940.

420. Eggleton, W. G. E. The zinc content of epidermal structures in beriberi. Biochem. J. 33:403-406, 1939.

421. Eilers, H. Zur Kenntnis der Ernährungsphysiologie von Stichococcus bacillaris Näg. Recueil Trav. Bot. Neerland. 23:362-395, 1926.

422. Eisenbrand, J., and M. Sienz. Über den Zinkgehalt von menschlichen Pankreasdrüsen. Hoppe-Seyler's Z. Physiol. Chem. 268:1-25, 1941.

422a. Ekberg, M., J.-O. Jeppsson, and T. Denneberg. Penicillamine treatment of cystinuria. Acta Med. Scand. 195:415-419, 1974.

423. El Arini, A. F., M. N. Salama, A. H. El Beheiry, G. El Din Massoud, and M. M. El Din Nofal. A new approach for evolution of the spermatogenic function using radiozinc. Fertil. Steril. 23:879-885, 1972.

423a. Elcoate, P. V., M. I. Fischer, C. A. Mawson, and M. J. Millar. The effect of zinc deficiency on the male genital system. J. Physiol. (Lond.) 129:53P-54P, 1955.

424. Elderfield, H., L. Thornton, and J. S. Webb. Heavy metals and oyster culture in Wales. Marine Pollut. Bull. 2:44-47, 1971.

425. Elgawhary, S. M., W. L. Lindsay, and W. D. Kemper. Effect of complexing agents and acids on the diffusion of zinc to a simulated root. Soil Sci. Soc. Amer. Proc. 34:211-214, 1970.

426. Elgawhary, S. M., W. L. Lindsay, and W. D. Kemper. Effect of EDTA on the self-diffusion of zinc in aqueous solution and in soil. Soil Sci. Soc. Amer. Proc. 34:66-70, 1970.

427. Elias, S., and M. Chvapil. Zinc and wound healing in normal and chronically ill rats. J. Surg. Res. 15:59-66, 1973.

427a. Eliasson, R., Ø. Johnsen, and C. Lindholmer. Effect of zinc on human sperm respiration. Life Sci. (Part I) 10:1317-1320, 1971.

428. Elkins, H. B. Zinc in air. Mixed-color dithizone method, pp. 414-416. In The Chemistry of Industrial Toxicology (2nd ed.). New York: John Wiley & Sons, Inc., 1959.

429. Elliot, J. S., and E. Eusebio. Calcium oxalate solubility: The effect of trace metals. Invest. Urol. 4:428-430, 1967.

430. Elliot, J. S., and M. E. Ribeiro. The urinary excretion of trace metals in patients with calcium oxalate urinary stone. Invest. Urol. 10: 253-255, 1973.

431. Ellis, B. G. Zinc deficiency. A symposium. Response and susceptibility. Crops Soils 18(1):13, 1965.

432. Ellis, B. G., and B. D. Knezek. Adsorption reactions of micronutrients in soils, pp. 59-78. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of Symposium, Muscle Shoals, Alabama, 1971. Madison, Wis.: Soil Science Society of America, 1972.

433. Ellis, E. H., J. A. Spadaro, and R. O. Becker. Trace elements in tendon collagen. Clin. Orthop. 65:195-198, 1969.

434. Ellis, J. H., R. I. Barnhisel, and R. E. Phillips. The diffusion of copper, manganese, and zinc as affected by concentration, clay mineralogy, and associated anions. Soil Sci. Soc. Amer. Proc. 34:866-870, 1970.

435. Ellis, R., Jr., J. F. Davis, and D. L. Thurlow. Zinc availability in calcareous Michigan soils as influenced by phosphorus level and temperature. Soil Sci. Soc. Amer. Proc. 28:83-86, 1964.

435a. Emanuel, M. B., and R. E. Oakey. Effect of Zn^{++} on the binding of oestradial-17β to a uterine protein. Nature 223:66-67, 1969.

436. Emes, J. H., and D. Arthur. The site of zinc absorption in the rat intestine. Proc. Soc. Exp. Biol. Med. 148:86-88, 1975.

436a. Eminians, J., J. G. Reinhold, G. A. Kfoury, G. H. Amirhakimi, H. Sharif, and M. Ziai. Zinc nutrition in children in Fars Province in Iran. Amer. J. Clin. Nutr. 20:734-742, 1967.

437. Engel, R. W., R. F. Miller, and N. O. Price. Metabolic patterns in pre-adolescent children: XIII. Zinc balance, pp. 326-337. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

438. English, R. M. The role of zinc in human nutrition. Food Nutr. Notes Rev. 26:45-56, 1969.

439. Englund, H. M., and W. T. Beery, Eds. Proceedings of the Second International Clean Air Congress. Held at Washington, D. C., Dec. 6-11, 1970. New York: Academic Press, 1971. 1354 pp.

440. Engström, L. Studies on calf-intestinal alkaline phosphatase. I. Chromatographic purification, microheterogeneity and some other properties of the purified enzyme. Biochim. Biophys. Acta 52:36-48, 1961.

440a. Environmental levels of radioactivity at Atomic Energy Commissions Installations. 1. Hanford atomic products operation, January-December, 1970. Radiat. Data Rep. 15:203-225, 1974.

440b. Entwisle, B. R. Acrodermatitis enteropathica. Report of a case in a twin with dramatic response to expressed human milk. Austral. J. Derm. 8:13-21, 1965.

441. Epstein, E., and J. E. Leggett. The absorption of alkaline earth cations by barley roots: Kinetics and mechanism. Amer. J. Bot. 41:785-791, 1954.

442. Erdman, J. W., Jr., and P. A. Lachance. Effect of salt mixture and cholesterol upon rat serum and liver zinc, vitamin A, and cholesterol. Nutr. Rep. Int. 9:319-329, 1974.

443. Ernst, W. Der Einfluss der Phosphatversorgung sowie die Werkung von ionogenem und chelatisiertem Zink auf die Zink- und Phosphataufnahme einiger Schwermetallpflanzen. Physiol. Plant. 21:323-333, 1968.

444. Ernst, W. Zur Kenntnis der Soziologie und Ökologie der Schwermetallvegetation Grossbritanniens. Ber. Dtsch. Bot. Ges. 81:116-124, 1968.

445. Evans, C. E., J. I. Wear, B. F. Hajek, and J. T. Cope, Jr. The relationship of soil zinc removed by three extractants to zinc uptake by corn and sorghum in medium- to fine-textured soils. Commun. Soil Sci. Plant Anal. 5:105-113, 1974.

446. Evans, D. W., and N. H. Cutshall. Effects of ocean water on the soluble suspended distribution of Columbia River radionuclides, pp. 125-140. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA, in Seattle, U.S.A., 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

447. Evans, G. W., C. I. Grace, and C. Hahn. Homeostatic regulation of zinc absorption in the rat. Proc. Soc. Exp. Biol. Med. 143:723-725, 1973.

448. Evans, G. W., C. I. Grace, and C. Hahn. The effect of copper and cadmium on ^{65}Zn absorption in zinc-deficient and zinc-supplemented rats. Bioinorg. Chem. 3:115-120, 1974.

449. Evans, G. W., C. I. Grace, and H. J. Votava. A proposed mechanism for zinc absorption in the rat. Amer. J. Physiol. 228:501-505, 1975.

450. Everson, R. G. Carbonic anhydrase and CO_2 fixation in isolated chloroplasts. Phytochemistry 9:25-32, 1970.

451. Fair, J. R., B. B. Crocker, and H. R. Null. Sampling and analyzing trace quantities. Chem. Eng. 79(21):146-154, 1972.

451a. Fairhall, L. T. Zinc, pp. 134-135. In Industrial Toxicology. (2nd ed.) Baltimore: Williams & Wilkins Company, 1957.

452. Falchuk, K. H., D. W. Fawcett, and B. W. Vallee. Role of zinc in cell division of Euglena gracilis. J. Cell Sci. 17:57-78, 1975.

453. Falin, L. I., and K. E. Gromzewa. Experimental teratoma testis in fowl produced by injections of zinc sulphate solution. Amer. J. Cancer 36:233-236, 1939.

453a. Favier, M., M. Yacoub, C. Racinet, C. Marka, P. Chabert, and A. Benbassa. Les ions métalliques dans le liquide amniotique au cours du troisième trimestre de la gestation. Relation significative entre la concentration en zinc et le poids foetal. Rev. Fr. Gynecol. 67:707-714, 1972.

454. Fazzari, C., and C. Catini. Alcuni aspetti morfologici dell'epitelio renale in corso di trattamento con cloruro di zinco. Sperimentale 117:299-314, 1967.

455. Feaster, J. P., S. L. Hansard, J. T. McCall, and G. K. Davis. Absorption, deposition and placental transfer of zinc^{65} in the rat. Amer. J. Physiol. 181:287-290, 1955.

456. Feaster, J. P., S. L. Hansard, J. T. McCall, F. H. Skipper, and G. K. Davis. Absorption and tissue distribution of radiozinc in steers fed high-zinc rations. J. Anim. Sci. 13:781-788, 1954.

457. Feaster, J. P., C. H. Van Middelem, and G. K. Davis. Zinc-DDT interrelationships in growth and reproduction in the rat. J. Nutr. 102: 523-528, 1972.

458. Feder, J., and L. R. Garrett. A rapid method for removal of zinc from the metallo neutral proteases. Biochem. Biophys. Res. Commun. 43: 943-948, 1971.

459. Federal Water Pollution Control Administration. Water Quality Criteria. Report of the National Technical Advisory Committee to the Secretary of the Interior April 1, 1968. Washington, D. C.: U. S. Government Printing Office, 1968. 234 pp.

460. Deleted.

460a. Felig, P., W. V. Brown, A. Levine, and G. Klatskin. Glucose homeostasis in viral hepatitis. New Engl. J. Med. 283:1436-1440, 1970.

461. Fell, B. F., L. C. Leigh, and R. B. Williams. The cytology of various organs in zinc-deficient rats with particular reference to the frequency of cell division. Res. Vet. Sci. 14:317-325, 1973.

462. Fell, G. S., D. P. Cuthbertson, C. Morrison, A. Fleck, K. Queen, R. G. Bessent, and S. L. Husain. Urinary zinc levels as an indication of muscle catabolism. Lancet 1:280-282, 1973.

463. Ferguson, H. W., and A. G. Leaver. Effects of high dietary zinc on the bones and teeth of rats maintained on low calcium regimes. J. Bone Joint Surg. 51 B:383-384, 1969. (abstract)

464. Ferguson, H. W., and A. G. Leaver. The effects of diets high in zinc at different levels of calcium and vitamin D on the rat humerus and incisor. Calcif. Tissue Res. 8:265-275, 1972.

465. Ferm, V. H., and S. J. Carpenter. Teratogenic effect of cadmium and its inhibition by zinc. Nature 216:1123, 1967.

466. Ferm, V. H., and S. J. Carpenter. The relationship of cadmium and zinc in experimental mammalian teratogenesis. Lab. Invest. 18:429-432, 1968.

467. Fernandez-Madrid, F., A. S. Prasad, and D. Oberleas. Effect of zinc deficiency on nucleic acids, collagen, and noncollagenous protein of connective tissue. J. Lab. Clin. Med. 82:951-961, 1973.

468. Ferrell, R. E., T. E. Carville, and J. D. Martinez. Trace metals in oyster shells. Environ. Lett. 4:311-316, 1973.

469. Feuerstein, H. Bestimmung von Cu, Ag, Au, Zn und As in menschlicher Haut mit Hilfe der Neutronenaktivierungsanalyse. Fresenius' Z. Anal. Chem. 232:196-197, 1967.

470. Fevold, H. L., F. L. Hisaw, and R. Greep. Augmentation of the gonad stimulating action of pituitary extracts by inorganic substances, particularly copper salts. Amer. J. Physiol. 117:68-74, 1936.

471. Filer, L. J. Excessive intake and imbalance of vitamins and minerals. Symposium on Vitamins and Minerals in Processed Foods, Council on Foods and Nutrition, AMA, and AMA Food Industry Liaison Committee, New Orleans, 1971.

472. Filo, R. S., C. H. Sloan, L. Weatherbee, and W. J. Fry. Influence of zinc and copper on the development of experimental atherosclerosis, pp. 476-481. In R. J. Jones, Ed. Proceedings of the Second International Symposium on Atherosclerosis held in Chicago, Illinois, Nov. 2-5, 1969. New York: Springer-Verlag, 1970.

473. Finelli, V. N., L. Murthy, W. B. Peirano, and H. G. Petering. δ-amino-levulinate dehydratase, a zinc dependent enzyme. Biochem. Biophys. Res. Commun. 60:1418-1424, 1974.

474. Fischer, G. M., E. I. Mata, and J. G. Llaurado. Regional differences in magnesium, calcium and zinc composition of arterial wall in normal and hypertensive dogs. Amer. Heart J. 75:784-789, 1968.

474a. Fischer, G. L., V. S. Byers, M. Shifrine, and A. S. Levin. Copper and zinc levels in serum from human patients with sarcomas. Cancer 37:356-363, 1976.

475. Fitzgerald, B. W., and D. M. Skauen. Zinc-65 in oysters in Fishers Island Sound and its estuaries, pp. 159-162. In V. Schultz and A. W. Klement, Jr., Eds. Radioecology. New York: Reinhold, 1963.

476. Fitzpatrick, T. B., M. Miyamoto, and K. Ishikawa. The evolution of concepts of melanin biology. Arch. Dermatol. 96:305-323, 1967.

477. Flanagan, F. J. U. S. Geological Survey Standards --II. First compilation of data for the new U. S. G. S. rocks. Geochim. Cosmochim. Acta 33: 81-120, 1969.

478. Fleischer, M. U. S. Geological Survey Standards -- I. Additional data on rocks G-1 and W-1, 1965-1967. Geochim. Cosmochim. Acta 33:65-79, 1969.

478a. Flitman, R., and Worth, Jr. Inhibition of hepatic alcohol dehydrogenase by bilirubin. J. Biol. Chem. 241:669-672, 1966.

479. Florence, E., and J. Quarterman. The effects of age, feeding pattern and sucrose on glucose tolerance, and plasma free fatty acids and insulin concentrations in rat. Brit. J. Nutr. 28:63-74, 1972.

479a. Flynn, A., W. J. Pories, W. H. Strain, and V. A. Hill, Jr. Zinc deficiency with altered adrenocortical function and its relation to delayed healing. Lancet 1:789-791, 1973.

480. Fogel, P. I. Interrelations between excretion of sex hormones and the copper, manganese, zinc and cobalt content in the placentas of women with a normal course and premature interruption of pregnancy. Akush. Ginekol. 47 (8):45-48, 1971. (in Russian, summary in English)

481. Falin, L. I., and K. E. Gromzeva. The pathogenesis of experimental teratoid tumors of genital glands. I. Experimental $ZnSO_4$ teratomas of the testicle of roosters. Arch. Sci. Biol. (Leningrad) 56(3): 101-111, 1939. (in Russian)

482. Foley, B., S. A. Johnson, B. M. Hackley, J. C. Smith, Jr., and J. A. Halsted. Zinc content of human platelets. Proc. Soc. Exp. Biol. Med. 128:265-269, 1968.

483. Falin, L. I., and K. E. Gromzeva. Pathogenesis of experimental teratoid tumors of genital glands. II. Teratoid tumors of the testes of roosters induced by injection of zinc nitrate solution. Arch. Sci. Biol. (Leningrad) 60(3):86-92, 1940. (in Russian)

484. Folk, J. E. Carboxypeptidase B., pp. 57-79. In P. D. Boyer, Ed. The Enzymes. Vol. 3. Hydrolysis: Peptide Bond. (3rd ed.) New York: Academic Press, 1971.

485. Folk, J. E., and J. A. Gladner. Cobalt activation of carboxypeptidase A. J. Biol. Chem. 235:60-63, 1960.

486. Folk, J. E., and E. W. Schirmer. The porcine pancreatic carboxypeptidase A system. I. Three froms of the active enzyme. J. Biol. Chem. 238:3884-3894, 1963.

487. Follett, R. H., and W. L. Lindsay. Profile Distribution of Zinc, Iron, Magnesium, and Copper in Colorado Soils. Technical Bulletin 110. Fort Collins: Colorado State University Experiment Station, 1970. 79 pp.

488. Follis, R. H., Jr. The pathology of zinc deficiency, pp. 129-141. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

488a. Follis, R. H., Jr., H. G. Day, and E. V. McCollum. Histological studies of the tissues of rats fed a diet extremely low in zinc. J. Nutr. 22:223-237, 1941.

489. Forbes, R. J. Zinc and brass, pp. 272-289. In Metallurgy in Antiquity. A Notebook for Archaeologists and Technologists. Leiden, Netherlands: E. J. Brill, 1950.

489a. Forbes, R. M. Mineral utilization in the rat. III. Effects of calcium, phosphorus, lactose and source of protein in zinc-deficient and in zinc-adequate diets. J. Nutr. 83:225-233, 1964.

490. Forman, H. J., H. J. Evans, R. L. Hill, and I. Fridovich. Histidine at the active site of superoxide dismutase. Biochemistry 12:823-827, 1973.

491. Forssén, A. Inorganic elements in the human body. I. Occurrence of Ba, Br, Ca, Cd, Cs, Cu, K, Mn, Ni, Sn, Sr, Y, and Zn in the human body. Ann. Med. Exp. Biol. Fenn. 50:99-162, 1972.

492. Forster, W. O. Radioactive and stable nuclides in the Columbia River and adjacent northeast Pacific Ocean, pp. 663-700. In A. T. Pruter and D. L. Alverson, Eds. The Columbia River Estuary and Adjacent Ocean Waters. Bioenvironmental Studies. Seattle: University of Washington Press, 1972.

493. Fosmire, G. J., and Y. Y. Al-Ubaidi, E. Halas, and H. H. Sandstead. The effect of zinc deprivation on the brain. Adv. Exp. Med. Biol. 48:329-345, 1974.

494. Fosmire, G. J., Y. Y. Al-Ubaidi, and H. H. Sandstead. Some effects of post-natal zinc deficiency on developing rat brain. Ped. Res. 9:89-93, 1975.

495. Fosmire, M. A., G. J. Fosmire, and H. H. Sandstead. Effects of zinc deficiency on polysomes. Fed. Proc. 33:699, 1974. (abstract)

496. Fosmire, G. J., and H. H. Sandstead. Alterations in protein synthesis in heart, liver, and kidney of zinc deficient suckling rat pups. Fed. Proc. 34:941, 1975. (abstract)

497. Foster, J. W. Chemical Activities of Fungi. New York: Academic Press, Inc., 1949. 648 pp.

498. Foster, J. W., and F. W. Denison, Jr. Role of zinc in metabolism. Nature 166:833-834, 1950.

499. Foster, R. F. The history of Hanford and its contribution of radionuclides to the Columbia River, pp. 3-18. In A. T. Pruter and D. L. Alverson, Eds. The Columbia River Estuary and Adjacent Ocean Waters. Bioenvironmental Studies. Seattle: University of Washington Press, 1972.

500. Fournier, P., and A. Digaud. Effets chez le rat de l'ingestion simultanée de lactose et de ^{65}Zn sur l'absorption et le retention de cet élément. C. R. Acad. Sci. D (Paris) 269:2001-2003, 1969.

501. Fowler, S. W., L. F. Small, and J. M. Dean. Distribution of ingested zinc-65 in the tissues of some marine crustaceans. J. Fish. Res. Board Can. 27:1051-1055, 1970.

502. Fowler, S. W., L. F. Small, and J. M. Dean. Experimental studies on elimination of zinc-65, cesium-137 and cerium-144 by euphausiids. Marine Biol. 8:224-231, 1971.

503. Fowler, S. W., L. F. Small, and J. M. Dean. Metabolism of zinc-65 in euphausiids, pp. 399-411. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

504. Fox, M. R. S. The status of zinc in human nutrition. World Rev. Nutr. Diet. 12:208-226, 1970.

504a. Fox, M. R. S., B. E. Fry, Jr., B. F. Harland, M. E. Schertel, and C. E. Weeks. Effect of ascorbic acid on cadmium toxicity in the young coturnix. J. Nutr. 101:1295-1305, 1971.

504b. Fredricks, R. E., K. R. Tanaka, and W. N. Valentine. Zinc in human blood cells: Normal values and abnormalities associated with liver disease. J. Clin. Invest. 39:1651-1656, 1960.

504c. Freier, S., J. Faber, R. Goldstein, and M. Mayer. Treatment of acrodermatitis enteropathica by intravenous amino acid hydrolysate. J. Pediatr. 82:109-112, 1973.

505. Fredricks, R. E., K. R. Tanaka, and W. N. Valentine. Variations of human blood cell zinc in disease. J. Clin. Invest. 43:304-314, 1964.

505a. Friberg, L., T. Kjellstrom, G. Nordberg, and M. Piscator. Cadmium in the Environment. III. A Toxicological and Epidemiological Appraisal. EPA-650/2-75-049. Stockholm, Sweden: The Karolinska Institute, 1975. 218 pp.

506. Friberg, L., M. Piscator, and G. Nordberg. Cadmium in the Environment. A Toxicological and Epidemiological Appraisal. APTD-0681. Stockholm: The Karolinska Institute. Department of Environmental Hygiene, 1971.

507. Friberg, L., M. Piscator, G. Nordberg, and T. Kjellström. Cadmium in the Environment, II. (Prepared for U. S. Environmental Protection Agency, Publ. EPA R2-73-190) Stockholm: The Karolinska Institute, Department of Environmental Hygiene, 1973. 121 pp.

508. Fridovich, I. Superoxide radical and superoxide dismutase. Accts. Chem. Res. 5:321-326, 1972.

509. Friedrichs, K.-H., and Y. P. Grover. The determination of heavy metals in dust samples from the atmospheric air by means of the ring oven method. Staub-Reinhalt. Luft (English Ed.) 32(1):30-34, 1972.

510. Frischauf, H., H. Altmann, and G. Stehlik. Investigations on the trace element content of leucocytes by means of neutron activation analysis, pp. 234-239. In Proceedings of the Ninth Congress of the European Society of Haematology, Lisboa 1963. Part II/1. New York: S. Karger, 1963.

511. Fritz, J. C. Effect of processing on the availability and nutritional value of trace mineral elements, pp. 109-118. In National Research Council, Agricultural Board, Committee on Animal Nutrition. Effect of Processing on the Nutritional Value of Feeds. Proceedings of a Symposium, Gainesville, Florida, January 11-13, 1972. Washington, D. C.: National Academy of Sciences, 1973.

511a. Friberg, J., and O. Nilsson. The amount of zinc detected in washed human spermatozoa. Upsala J. Med. Sci. 79:63-64, 1974.

512. Fuge, R., and K. H. James. Trace metal concentrations in brown seaweeds. Cardigan Bay, Wales. Marine Chem. 1:281-293, 1973.

512a. Fukubayashi, H. H., and L. W. Higley. Recovery of Zinc and Lead from Brass Smelter Dust. Bureau of Mines Report of Investigations 7880. Washington, D. C.: U. S. Department of the Interior, 1974. 10 pp.

513. Fujii, T. Presence of zinc in nucleoli and chromosomes and its possible role in mitosis. J. Faculty Sci. Univ. Tokyo (Tokyo Daigaku Rigakubu) 7(Sect. 4):313-325, 1955.

514. Fujii, T., S. Utida, and T. Mizuno. Reaction of starfish spermatozoa to histidine and certain other substances considered in relation to zinc. Nature 176:1068-1069. 1955. (letter)

515. Fujioka, M., and I. Lieberman. A Zn^{++} requirement for synthesis of deoxyribonucleic acid by rat liver. J. Biol. Chem. 239:1164-1167, 1964.

515a. Furchner, J. E., and C. R. Richmond. Effect of dietary zinc on the absorption of orally administered Zn^{65}. Health Phys. 8:35-40, 1962.

516. Furst, A. Trace elements and cancer, pp. 14-26. In Chemistry of Chelation in Cancer. Springfield, Ill.: Charles C Thomas, 1963.

517. Furth, A. J. Purification and properties of horse erythrocyte carbonic anhydrases. J. Biol. Chem. 243:4832-4841, 1968.

518. Fuwa, K., W. E. C. Wacker, R. Druyan, A. F. Bartholomay, and B. L. Vallee. Nucleic acids and metals, II: Transition metals as determinants of the conformation of ribonucleic acids. Proc. Nat. Acad. Sci. U.S.A. 46:1298-1307, 1960.

518a. Gabbiani, G., D. Baic, and C. Déziel. Studies on tolerance and ionic antagonism for cadmium or mercury. Can J. Physiol. Pharmacol. 45: 443-450, 1967.

519. Gall, O. E., and R. M. Barnette. Toxic limits of replaceable zinc to corn and cowpeas grown on three Florida soils. J. Amer. Soc. Agron. 32:23-32, 1940.

520. Gallery, E. D. M., J. Blomfield, and S. R. Dixon. Acute zinc toxicity in haemodialysis. Brit. Med. J. 1972(4):331-333.

521. Galtsoff, P. S. Accumulation of manganese, iron, copper and zinc in the body of American oyster. Crassostrea (Ostrea) virginica. Anat. Rec. 117:601-602, 1953.

522. Galtsoff, P. S. Iron, copper, zinc and manganese, pp. 387-390. In The American Oyster, Crassostrea virginica Gmelin. U. S. Fishery Bulletin of the Fish and Wildlife Service. Vol. 64. Washington, D. C.: U. S. Government Printing Office, 1964.

523. Ganiron, R. B., D. C. Adriano, G. M. Paulsen, and L. S. Murphy. Effect of phosphorus carriers and zinc sources on phosphorus-zinc interaction in corn. Soil Sci. Soc. Amer. Proc. 33:306-309, 1969.

524. Gasaway, W. C., and I. O. Buss. Zinc toxicity in the Mallard duck. J. Wildlife Manage. 36:1107-1117, 1972.

525. Geering, H. R., and J. F. Hodgson. Micronutrient cation complexes in soil solution: III. Characterization of soil solution ligands and their complexes with Zn^{2+} and Cu^{2+}. Soil Sci. Soc. Amer. Proc. 33:54-59, 1969.

526. Geering, H. R., J. F. Hodgson, and C. Sdano. Micronutrient cation complexes in soil solution: IV. The chemical state of manganese in soil solution. Soil Sci. Soc. Amer. Proc. 33:81-85, 1969.

526a. Gerber, D. A. Decreased concentration of free histidine in serum in rheumatoid arthritis, an isolated amino acid abnormality not associated with general hypoaminoacidemia. J. Rheumatol. 2:384-392, 1975.

527. Gerloff, G. C. Plant Analysis for Nutrient Assay of Natural Waters. Office of Research and Monitoring Report EPA-R1-73-001. Washington, D. C.: U. S. Environmental Protection Agency, 1973. 66 pp.

528. Gershoff, S. N. Effects of vitamin B_6 and B_1 deficiencies and restricted caloric intake on tissue zinc level. Proc. Soc. Exp. Biol. Med. 127: 1207-1210, 1968.

529. Giauque, R. D., L. Y. Goda, and N. E. Brown. Characterization of aerosols in California by X-ray-induced X-ray fluorescence analysis. Environ. Sci. Technol. 8:436-441, 1974.

530. Gilbert, I. G. F., and D. M. Taylor. The behavior of zinc and radiozinc in the rat. Biochim. Biophys. Acta 21:545-551, 1956.

531. Gilfrich, J. V., P. G. Burkhalter, and L. S. Birks. X-ray spectrometry for particulate air pollution -- a quantitative comparison of techniques. Anal. Chem. 45:2002-2009, 1973.

532. Deleted

533. Ginsberg, D., and J. M. Hsu. Impairment of tryptophan-^{3}H incorporation into skin protein by zinc-deficient rats. Fed. Proc. 33:699, 1974. (abstract)

534. Giordano, P. M., and J. J. Mortvedt. Response of several rice cultivars to Zn. Agron. J. 66:220-223, 1974.

535. Giordano, P. M., and J. J. Mortvedt. Rice response to Zn in flooded and nonflooded soil. Agron. J. 64:521-524, 1972.

536. Giordano, P. M., J. C. Noggle, and J. J. Mortvedt. Zinc uptake by rice, as affected by metabolic inhibitors and competing cations. Plant Soil 41: 637-646, 1974.

537. Giroux, E. L., and R. I. Henkin. Competition for zinc among serum albumin and amino acids. Biochim. Biophys. Acta 273:64-72, 1972.

538. Giroux, E. L., and R. I. Henkin. Macromolecular ligands of exchangeable copper, zinc and cadmium in human serum. Bioinorg. Chem. 2:125-233, 1972.

539. Gladney, E. S., W. H. Zoller, A. G. Jones, and G. E. Gordon. Composition and size distributions of atmospheric particulate matter in Boston area. Environ. Sci. Technol. 8:551-557, 1974.

540. Gladstones, J. S., and J. F. Loneragen. Mineral elements in temperate crops and pasture plants. I. Zinc. Austral. J. Agric. Res. 18: 427-446, 1967.

541. Gleit, C. E., and W. D. Holland. Electrically excited oxygen for the low temperature decomposition of organic substances. Anal. Chem. 34:1454-1457, 1962.

541a. Glossmann, H., and D. M. Neville, Jr. Phlorizin receptors in isolated kidney brush border membranes. J. Biol. Chem. 247:7779-7789, 1972.

542. Gofman, J. W., O. F. deLalla, E. I. Kovich, O. Lowe, W. Martin, D. L. Piluso, R. K. Tandy, and F. Upham. Chemical elements of the blood of man. Arch. Environ. Health 8:105-109, 1964.

542a. Goldberg, A. L., and D. F. Goldspink. Influence of food deprivation and adrenal steroids on DNA synthesis in various mammalian tissues. Amer. J. Physiol. 228:310-317, 1975.

543. Goldberg, E. D. Minor elements in sea water, pp. 163-196. In J. P. Riley and G. Skirrow, Eds. Chemical Oceanography. Vol. 1. New York: Academic Press, 1965.

544. Gombe, S., J. Apgar, and W. Hansel. Effect of zinc deficiency and restricted food intake on plasma and pituitary LH and hypothalamic LRF in female rats. Biol. Reprod. 9:415-419, 1973.

544a. Gonick, P., D. Oberleas, T. Knechtges, and A. S. Prasad. Atomic absorption spectrophotometric determination of zinc in the prostate. Invest. Urol. 6:345-347. 1969.

545. Gontzea, I., and P. Sutzescu. Table VI. Phytic-acid contents in foodstuffs, pp. 58-59. In Natural Antinutritive Substances in Foodstuffs and Forages. New York: S. Karger, 1968.

545a. Gooden, C. S. Non-destructive neutron activiation analysis for the determination of manganese and zinc in human skin biopsies. Phys. Med. Biol. 17:26-31, 1972.

546. Goodman, G. T., and T. M. Roberts. Plants and soils as indicators of metals in the air. Nature 231:287-292, 1971.

547. Gordon, G. E. Instrumental activation analysis of atmospheric pollutants and pollution source materials, pp. 138-143. In B. Westley, Ed. Proceedings of the International Symposium on Identification and Measurement of Environmental Pollutants. Ottawa, Ontario, Canada, June 14-17, 1971. Ottawa: National Research Council of Canada, 1971.

548. Gordon, G. E., W. H. Zoller, and E. S. Gladney. Abnormally enriched trace elements in the atmosphere, pp. 167-174. In D. C. Hemphill, Ed. Trace Substance in Environmental Health - VII. Proceedings of 7th Annual Conference held in Columbia, Missouri, June 12-14, 1973. Columbia: University of Missouri, 1973.

549. Gormican, A. Inorganic elements in foods used in hospital menus. J. Amer. Diet. Assoc. 56:397-403, 1970.

550. Gorsline, G. W., D. E. Baker, and W. I. Thomas. Accumulation of Eleven Elements by Field Corn (Zea mays L.) Pennsylvania Agricultural Experiment Station Bulletin 725. University Park: Pennsylvania State University, College of Agriculture, 1965. 34 pp.

551. Gorsuch, T. T. The Destruction of Organic Matter. New York: Pergamon Press, 1970. 152 pp.

552. Grace, N. D. Observations on plasma zinc levels in sheep grazing New Zealand pastures. N. Z. J. Agric. Res. 15:284-288, 1972.

553. Gracy, R. W., and E. A. Noltmann. Studies on phosphomannose isomerase. II. Characterization as a zinc metalloenzyme. J. Biol. Chem. 243: 4109-4116, 1968.

554. Graham, W. M. S. On the internal use of sulphate of zinc in gleet and leucorrhea. Edinburgh Med. Surg. J. 26:*107-*108, 1826.

554a. Graham, D., and M. L. Reed. Carbonic anhydrase and the regulation of photosynthesis. Nature New Biol. 231:81-83, 1971.

555. Graham, R., J. Sampson, and H. R. Hester. Results of feeding zinc to pregnant mares and to mares nursing foals. J. Amer. Vet. Med. Assoc. 97:41-47, 1940.

556. Grant-Frost, D. R., and E. J. Underwood. Zinc toxicity in the rat and its interrelation with copper. Austral. J. Exp. Biol. Med. Sci. 36:339-346, 1958.

557. Gravens, D. L., H. W. Margraf, H. R. Butcher, Jr., and W. F. Ballinger. The antibacterial effect of treating sutures with silver. Surgery 73:122-127, 1973.

558. Gray, D., D. M. McKown, M. Kay, M. Eichor, and J. R. Vogt. Determination of trace element levels in atmospheric pollutants by instrumental neutron activation analysis. IEEE Trans. Nucl. Sci. 19(1):194-198, 1972.

559. Greaves, M., and T. R. C. Boyde. Plasma-zinc concentrations in patients with psoriasis, other dermatoses and venous leg ulceration. Lancet 2:1019-1020, 1967.

559a. Greaves, M. W., and F. A. Ive. Double-blind trial of zinc sulphate in the treatment of chronic venous leg ulceration. Brit. J. Dermatol. 87: 632-634, 1972.

560. Greaves, M. W., and A. W. Skillen. Effects of long-continued ingestion of zinc sulphate in patients with venous leg ulceration. Lancet 2:889-891, 1970.

561. Deleted.

562. Greene, H. L., G. Merenstein, M. Hambidge, H. E. Sauberlich, and Y. F. Herman. Trace elements and vitamins (V) in total parenteral nutrition (TPN). Pediatr. Res. 7:337, 1973. (abstract)

562a. Delete -- use 526a.

563. Gregory, R. P. G., and A. D. Bradshaw. Heavy metal tolerance in populations of Agrostis tenuis, Sibth, and other grasses. New Phytol. 64:131-143, 1965.

564. Greifer, B., and J. K. Taylor. Survey of Various Approaches to the Chemical Analysis of Environmentally Important Materials. National Bureau of Standards Internal Report 73-209. Prepared for the Environmental Protection Agency, Research Triangle Park, N. C., July 1973. 226 pp.

565. Greszta, J., and S. Godzik. Effect of zinc metallurgy on soils. Rocz. Gleboznawcze 20:195-215, 1969. (in Polish, summary in English)

566. Griffith, K., E. B. Wright, and T. L. Dormandy. Tissue zinc in malignant disease. Nature 241:60, 1973.

567. Griffith, P. R., and J. C. Alexander. Effect of zinc deficiency on amino acid metabolism of the rat. Nutr. Rep. Int. 6:9-20, 1972.

568. Grimmett, R. E. R., I. G. McIntosh, E. M. Wall, and C. S. M. Hopkirk. Chronic zinc-poisoning of pigs. Results of experimental feeding of pure zinc lactate. N. Z. J. Agric. 54:216-223, 1937.

569. Groppel, B., and H. Hennig. Zinkmangel beim Wiederkäuer. Arch. Exp. Veterinärmed. 25:817-821, 1971.

570. Gross, M. G., and C. A. Barnes. Radioactivity of the Columbia River effluent. Science 149:1088-1090, 1965.

571. Groundwater, W., and I. B. MacLeod. The effects of systemic zinc supplements on the strength of healing incised wounds in normal rats. Brit. J. Surg. 57:222-225, 1970.

571a. Henkin, R. I., and W. D. Grover. Trichopoliodystrophy (TPD): A genetic defect in selective Cu binding and storage as well as transport. Clin. Res. 24:435A, 1976. (abstract)

572. Grunes, D. L., L. C. Boawn, C. W. Carlson, and F. G. Viets, Jr. Zinc deficiency of corn and potatoes as related to soil and plant analyses. Agron. J. 53:68-67, 1961.

573. Guggenheim, K. The role of zinc, copper and calcium in the etiology of the "meat anemia." Blood 23:786-794, 1964.

574. Guinn, G., and H. E. Joham. Effects of two chelating agents on absorption and translocation of Fe, Cu, Mn, and Zn by the cotton plant. Soil Sci. 94:220-223, 1962.

575. Gulbrandsen, R. A. Chemical composition of phosphorites of the Phosphoria Formation. Geochim. Cosmochim. Acta 30:769-778, 1966.

575a. Gunn, S. A., and T. C. Gould. The role of zinc in the posttesticular antifertility action of monochlorhydrin. Proc. Soc. Exp. Biol. Med. 141:639-642, 1972.

575b. Gunn, S. A., and T. C. Gould. Cadmium and other mineral elements, pp. 378-481. In A. D. Johnson, W. R. Gomes and N. L. Van Demark, Eds. The Testis. Vol. 3. Influencing Factors. New York: Academic Press, 1970.

576. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. Cadmium-induced interstitial cell tumors in rats and mice and their prevention by zinc. J. Nat. Cancer Inst. 31:745-759, 1963.

577. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. Competition of cadmium for zinc in rat testis and dorsolateral prostate. Acta Endocrinol. 37:24-30, 1961.

578. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. Interference with fecal excretion of Zn^{65} by cadmium. Proc. Soc. Exp. Biol. Med. 111:559-562, 1962.

579. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. Mechanisms of zinc cysteine and selenium protection against cadmium-induced vascular injury to mouse testis. J. Reprod. Fertil. 15:65-70, 1968.

580. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. Prenatal and postnatal transfer of zinc-65. Radiat. Res. 20:504-509, 1963.

581. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. Selectivity of organ response to cadmium injury and various protective measures. J. Path. Bacteriol. 96:89-96, 1968.

582. Gunn, S. A., T. C. Gould, and W. A. D. Anderson. The selective injurious response of testicular and epididymal blood vessels to cadmium and its prevention by zinc. Amer. J. Path. 42:685-702, 1963.

583. Gunn, S. A., T. C. Gould, and W. A. Anderson. Zinc protection against cadmium injury to rat testis. Arch. Path. 71:274-281, 1961.

584. Gunn, S. A., T. C. Gould, S. S. Ginori, and J. G. Morse. Selective uptake of Zn^{65} by dorsolateral prostate of rat. Proc. Soc. Exp. Biol. Med. 88:556-558, 1955.

585. Gurba, P. E., R. E. Sennett, and R. D. Kobes. Studies on the mechanism of action of δ-aminolevulinate dehydratase from bovine and rat liver. Arch. Biochem. Biophys. 150:130-136, 1972.

586. Gurd, F. R. N., and D. S. Goodman. Preparation and properties of serum and plasma proteins. XXXII. The interaction of human serum albumin with zinc ions. J. Amer. Chem. Soc. 74:670-675, 1952.

587. Gurevich, G. P. Natural copper and zinc content in food products originating from Primorie territory. Vopr. Pitan. 20(5):38-40, 1961. (in Russian, summary in English)

587a. Gurney, J. J., and L. H. Ahrens. The zinc content of some ultramafic and basic rocks. Trans. Geol. Soc. S. Afr. 76:301-307, 1973.

588. Gustafson, P. F. Comments on radionuclides in aquatic ecosystems, pp. 853-858. In B. Åberg, and F. P. Hungate. Radioecological Concentration Processes. Proceedings of an International Symposium held in Stockholm, 25-29, April 1966. New York: Pergamon Press, 1967.

589. Guthrie, J. Zinc induction of testicular teratomas in Japanese quail (Coturnix coturnix japonica) after photoperiodic stimulation of testis. Brit. J. Cancer 25:311-314, 1971.

590. Gutknecht, J. Mechanism of radioactive zinc uptake by Ulva lactuca. Limnol. Oceanogr. 6:426-231, 1961.

591. Gutknecht, J. Uptake and retention of cesium 137 and zinc 65 by seaweeds. Limnol. Oceanogr. 10:58-66, 1965.

592. Gutknecht, J. Zn^{65} uptake by benthic marine algae. Limnol. Oceanogr. 8:31-38, 1963.

593. Gyorkey, F., K.-W. Min, J. A. Huff, and P. Gyorkey. Zinc and magnesium in human prostate gland: Normal, hyperplastic and neoplastic. Cancer Res. 27:1348-1353, 1967.

594. Gyorkey, F., and C. S. Sato. In vitro ^{65}Zn-binding capacities of normal, hyperplastic and carcinomatous human prostate gland. Exp. Mol. Path. 8:216-224, 1968.

595. Haaranen, S. Some observations on the zinc requirement of cattle for the prevention of itch and hair licking at different calcium levels in the feed. Nord. Vet. Med. 15:536-542, 1963.

595a. Hackley, B. M., J. C. Smith, and J. A. Halsted. A simplified method for plasma zinc determination by atomic absorption spectrophotometry. Clin. Chem. 14:1-5, 1968.

596. Hagenfeldt, K., L. O. Plantin, and E. Diczfalusy. Trace elements in the human endometrium. 1. Zinc, copper, manganese, sodium and potassium concentrations at various phases of the normal menstrual cycle. Acta Endocrinol. 65:541-551, 1970.

597. Hagenfeldt, K., L. O. Plantin, and E. Diczfalusy. Trace elements in the human endometrium. 2. Zinc, copper and manganese levels in the endometrium, cervical mucus and plasma. Acta Endocrinol. 72:115-126, 1973.

598. Haghiri, F. Plant uptake of cadmium as influenced by cation exchange capacity, organic matter, zinc and soil temperature. J. Environ. Qual. 3:180-183, 1974.

598a. Kägi, J. H. R., and B. L. Vallee. The role of zinc in alcoholic dehydrogenase. V. The effect of metal-binding agents on the structure of the yeast alcohol dehydrogenase molecule. J. Biol. Chem. 235:3188-3192, 1960.

599. Hähn, N., K. Paschen, and J. Haller. Das Verhalten von Kupfer, Eisen, Magnesium, Calcium und Zink bei Frauen mit normalem Menstruationscyclus, unter Einnahme von Ovulationshemmern und in der Gravidität. Arch. Gynäk. 213: 176-186, 1972.

600. Halas, E. S., M. J. Hanlon, and H. H. Sandstead. Prenatal nutrition and aggression. Fed. Proc. 34:940, 1975. (abstract)

601. Halas, E. S., and H. H. Sandstead. Some effects of prenatal zinc deficiency on behavior of the adult rat. Pediatr. Res. 9:94-97, 1975.

602. Halford, S. E., D. A. Lennette, P. M. Kelley, and M. J. Schlesinger. A mutationally altered alkaline phosphatase from Escherichia coli. I. Formation of an active enzyme in vitro and phenotypic suppression in vivo. J. Biol. Chem. 247:2087-2094, 1972.

603. Halim, A. H., C. E. Wassom, and R. Ellis, Jr. Zinc deficiency symptoms and zinc and phosphorous interactions in several strains of corn (Zea mays L.). Agron. J. 60:267-271, 1968.

604. Hall, T. X-ray fluorescence analysis in biology. Science 134:449-455, 1961.

604a. Hall, T. A. The microprobe analysis of zinc in mammalian sperm cells, pp. 679-685. In R. Castaing, P. Deschamps, and J. Philibert, Eds. Optique des Rayons X et Microanalyse. IV Congrès International, Orsay, Septembre 1965. Paris: Hermann, 1966.

605. Hallbook, T., and E. Lanner. Serum-zinc and healing of venous leg ulcers. Lancet 2:780-782, 1972.

606. Hallman, P. S., D. D. Perrin, and A. E. Watt. The computed distribution of copper (II) and zinc(II) ions among seventeen amino acids present in human blood plasma. Biochem. J. 121:549-555, 1971.

606a. Hallowell, J. B., J. F. Shea, G. R. Smithson, Jr., A. B. Tripler, and B. W. Gonser. Water-Pollution Control in the Primary Nonferrous-Metals Industry. Vol. 1. Copper, Zinc, and Lead Industries. EPA-R2-73-247a. Washington, D. C.: U. S. Government Printing Office, 1973. 168 pp.

607. Halstead, E. H., S. A. Barber, D. D. Warncke, and J. B. Bole. Supply of Ca, Sr, Mn and Zn to plant roots growing in soil. Soil Sci. Soc. Amer. Proc. 32:69-72, 1968.

608. Delete 608--same as 613.

609. Halsted, J. A. Zinc deficiency in man. Lancet 1:1447-1448, 1963.

610. Halsted, J. A., B. M. Hackley, C. Rudzki, and J. C. Smith, Jr. Plasma zinc concentration in liver diseases. Comparison with normal controls and certain other chronic diseases. Gastroenterology 54: 1098-1105, 1968.

611. Halsted, J. A., B. M. Hackley, and J. C. Smith, Jr. Plasma-zinc and copper in pregnancy and after oral contraceptives. Lancet 2:278-279, 1968. (letter)

612. Halsted, J. A., H. A. Ronaghy, P. Abadi, M. Haghshenass, G. H. Amirhakemi, R. M. Barakat, and J. G. Reinhold. Zinc deficiency in man. The Shiraz experiment. Amer. J. Med. 53:277-284, 1972.

612a. Halsted, J. A., and J. C. Smith, Jr. Night blindness and chronic liver disease. Gastroenterology 67:193-194, 1974.

613. Halsted, J. A., and J. C. Smith, Jr. Plasma-zinc in health and disease. Lancet 1:322-324, 1970.

614. Halsted, J. A., J. C. Smith, Jr., and M. I. Irwin. A conspectus of research on zinc requirements of man. J. Nutr. 104:347-378, 1974.

614a. Hambidge, K. M. The clinical significance of trace element deficiencies in man. Proc. Nutr. Soc. 33:249-255, 1974.

614b. Delete--use 620a.

615. Hambidge, K. M. Zinc deficiency in children, pp. 171-183. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals--2. Proceedings of the 2nd International Symposium, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

616. Hambidge, K. M., and W. Droegemueller. Changes in plasma and hair concentrations of zinc, copper, chromium, and manganese during pregnancy. Obstet. Gynecol. 44:666-672, 1974.

617. Hambidge, K. M., C. Hambidge, M. Jacobs, and J. D. Baum. Low levels of zinc in hair, anorexia, poor growth, and hypogeusia in children. Pediatr. Res. 6:868-874, 1972.

617a. Delete

618. Hambidge, K. M., K. H. Neldner, and P. A. Walravens. Zinc, acrodermatitis enteropathica, and congenital malformations. Lancet 1:577-578, 1975.

619. Hambidge, K. M., K. H. Neldner, and P. A. Walravens. Zinc and acrodermatitis enteropathica. Pediatr. Res. 9:283, 1975. (abstract)

620. Hambidge, K. M., and D. O. Rodgerson. Comparison of hair chromium levels of nulliparous and parous women. Amer. J. Obstet. Gynecol. 103:320-321, 1969.

620a. Hambidge, K. M., and P. A. Walravens. Zinc deficiency in infants and preadolescent children, pp. 21-32. In A. S. Prasad and D. Oberleas, Eds. Trace Elements in Human Health and Disease. Vol. 1. Zinc and Copper. New York: Academic Press, 1976.

621. Hambidge, K. M., and A. Silverman. Pica with rapid improvement after dietary zinc supplementation. Arch. Dis. Child. 48:567-568, 1973.

621a. Hambidge, K. M., P. Walravens, V. Kumar, and C. Tuchinda. Chromium, zinc, manganese, copper, nickel, iron and cadmium concentrations in the hair of residents of Chandigarh, India and Bangkok, Thailand, pp. 39-44. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - VIII. Proceedings of University of Missouri's 8th Annual Conference held June 11-13, 1974. Columbia: University of Missouri, 1974.

621b. Hamburgh, M., M. Erlich, G. Nathanson, and I. Pesetsky. Additional observations relating to the mechanism of trypan blue induced teratogenesis. J. Exp. Zool. 192:1-12, 1975.

622. Hamdi, E. A. Chronic exposure to zinc of furnace operators in a brass foundry. Brit. J. Ind. Med. 26:126-134, 1969.

623. Hamilton, E. I., M. J. Minski, and J. J. Cleary. The concentration and distribution of some stable elements in healthy human tissues from the United Kingdom. Sci. Total Environ. 1:341-374, 1973.

624. Hamilton, E. I., M. J. Minski, and J. J. Cleary. The loss of elements during the decomposition of biological materials with special reference to arsenic, sodium, strontium and zinc. Analyst 92:257-259, 1967.

624a. Hammer, D. I., J. F. Finklea, R. H. Hendricks, C. M. Shy, and R. J. M. Horton. Hair trace metal levels and environmental exposure. Amer. J. Epidemiol. 93:84-92, 1971.

625. Hammerle, R. H., R. H. Rarsh, K. Rengan, R. D. Giauque, and J. M. Jaklevic. Text of x-ray fluorescence spectrometry as a method for analysis of the elemental composition of atmospheric aerosols. Anal. Chem. 45: 1939-1940, 1973.

625a. Hammond, J. B., H. R. Black, and J. L. Cullison. Effect of oral zinc therapy in cirrhosis. Amer. J. Dig. Dis. 5:923-930, 1960.

626. Hampton, D. L., W. J. Miller, D. M. Blackmon, R. P. Gentry, M. W. Neathery, and P. E. Stake. Intestinal sites of zinc absorption in intact male Holstein calves. Fed. Proc. 34:907, 1975. (abstract)

627. Hanig, R. C., and M. H. Aprison. Determination of calcium, copper, iron, magnesium, manganese, potassium, sodium, zinc and chloride concentrations in several brain areas. Anal. Biochem. 21:169-177, 1967.

628. Hanna, W. H. Methods for chemical analysis of soils, pp. 474-502. In F. E. Bear, Ed. Chemistry of the Soil. (2nd ed.) New York: Reinhold Publishing Corporation, 1964.

629. Hansard, S. L. Placental transfer and fetal utilization of absorbed minerals by developing swine, pp. 79-86. In L. K. Bustad, R. O. McClellan and M. P. Burns, Eds. Swine in Biomedical Research. Seattle: Frayn Printing Co., 1966.

630. Hansard, S. L. Transplacental movement and maternal-fetal organ accretion rates of selected radiominerals in gravid cattle, sheep and swine, pp. 9-23. In M. R. Sikov and D. D. Mahlum, Eds. Radiation Biology of the Fetal and Juvenile Mammal. Proceedings of the Ninth Annual Hanford Biology Symposium at Richland, Washington, May 5-8, 1969. CONF-690501. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

630a. Hansen, J. D. L., and B. H. Lehmann. Serum zinc and copper concentrations in children with protein-calorie malnutrition. S. Afr. Med. J. 43: 1248-1251, 1969.

631. Haq, A. U., and M. H. Miller. Prediction of available soil Zn, Cu, and Mn using chemical extractants. Agron. J. 64:779-782, 1972.

632. Harding, M. M., D. C. Hodgkin, A. F. Kennedy, A. O'Connor, and P. D. J. Weitzmann. The crystal structure of insulin. II. An investigation of rhombohedral zinc insulin crystals and a report of other crystalline forms. J. Mol. Biol. 16:212-226, 1966.

632a. Handjani, A. M., J. C. Smith, Jr., J. B. Herrmann, and J. A. Halsted. Serum zinc concentrations in actue myocardial infarction. Chest 65:185-187, 1974.

633. Harkness, D. R. Studies on human placental alkaline phosphatase. I. Purification and crystallization. Arch. Biochem. Biophys. 126:503-512, 1968.

634. Harland, B. F., M. R. S. Fox, and B. E. Fry, Jr. Changes in plasma zinc related to fasting and dietary protein intake of Japanese quail. Proc. Soc. Exp. Biol. Med. 145:316-322, 1974.

634a. Harland, B. F., B. E. Fry, Jr., R. M. Jacobs, A. O. Lee, and M. R. S. Fox. Dietary protein source and tissue zinc in young Japanese quail. Fed. Proc. 33:700, 1974. (abstract)

635. Haroz, R. K., J. S. Twu, and R. K. Bretthauer. Purification and properties of a yeast nucleotide pyrophosphatase. J. Biol. Chem. 247:1452-1457, 1972.

635a. Harris, A. B. Inhibition of growth and nucleic acid synthesis in zinc-deficient Mycobacterium smegmatis. J. Gen. Microbiol. 56:27-33, 1969.

636. Harris, I. Structure and catalytic activity of alcohol dehydrogenases. Nature 203:30-34, 1964.

637. Harrison, F. L. Accumulation and distribution of ^{54}Mn and ^{65}Zn in freshwater clams, pp. 198-220. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

638. Harrison, F. L., and D. J. Quinn. Tissue distribution of accumulated radionuclides in freshwater clams. Health Phys. 23:509-517, 1972.

639. Harrison, J. H. Participation of Zn^{++} in mechanism of action of malic dehydrogenase. Fed. Proc. 22:493, 1963. (abstract)

640. Harrison, P. R., K. A. Rahn, R. Dams, J. A. Robbins, J. W. Winchester, S. S. Brar, and D. M. Nelson. Areawide trace metal concentrations measured by multielement neutron activation analysis. A one day study in Northwest Indiana. J. Air. Pollut. Control Assoc. 21:563-570, 1971.

641. Harrison, W. W., M. G. Netsky, and M. D. Brown. Trace elements in human brain: Copper, zinc, iron and magnesium. Clin. Chim. Acta 21:55-60, 1968.

642. Harrison, W. W., J. P. Yurachek, and C. A. Benson. The determination of trace elements in human hair by atomic absorption spectroscopy. Clin. Chim. Acta 23:83-91, 1969.

643. Hartley, T. F., J. B. Dawson, and A. Hodgkinson. Simultaneous measurement of sodium, potassium, calcium, magnesium, copper and zinc balances in man. Clin. Chim. Acta 52:321-333, 1974.

644. Hartsuck, J. A., and W. N. Lipscomb. Carboxypeptidase A, pp. 1-56. In P. D. Boyer, Ed. The Enzymes. Vol. 3. Hydrolysis: Peptide Bond. (3rd ed.) New York: Academic Press, 1971.

645. Hartz, F. W., and H. F. Deutsch. Subunit structure of human superoxide dismutase. J. Biol. Chem. 247:7043-7050, 1972.

646. Harvey, H. W. The Chemistry and Fertility of Sea Waters. Cambridge: Cambridge University Press, 1955. 224 pp.

647. Harvey, R. S. Effects of temperature on the absorption of radionuclides by a blue-green alga, pp. 266-269. In D. J. Nelson, and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

648. Harvey, R. S. Uptake and loss of radionuclides by the freshwater clam Lampsilis radiata (Gmel.). Health Phys. 17:149-154, 1969.

649. Harvey, R. S., and R. Patrick. Concentration of ^{137}Cs, ^{65}Zn and ^{85}Sr by fresh-water algae. Biotechnol. Bioeng. 9:449-456, 1967.

650. Hauck, G. Erfahrungen mit der flammenlosen Atomabsorption bei der Untersuchung biologishen Materials auf Spuren von Schwermetallen. Fresenius' Z. Anal. Chem. 267:337-341, 1972.

651. Haumont, S., and F. C. McLean. Zinc and physiology of bone, pp. 169-186. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

652. Hawkins, G. W., D. C. Martens, and G. D. McCart. Response of corn to plowed-down and disked-in Zn as zinc sulfate. Commun. Soil Sci. Plant. Anal. 4:407-412, 1973.

653. Hawley, J. E. Spectrographic study of some Nova Scotia coals. Trans. Can. Inst. Mining Metall. 58:412-426, 1955.

654. Hayman, S., and E. K. Patterson. Purification and properties of a mouse ascites tumor dipeptidase, a metalloenzyme. J. Biol. Chem. 246: 660-669, 1971.

655. Headlee, A. J. W., and R. G. Hunter. The inorganic elements in the coals, pp. 36-122. In West Virginia Geological Survey (Reports) Vol. XIII A. Suppl. Part V., 1955.

656. Healy, W. B. Influence of soil type on ingestion of soil by grazing animals, pp. 437-445. In 9th International Congress of Soil Science Transactions, Adelaide, Australia, 1968. Vol. III. New York: American Elsevier Publishing Co., Inc., 1968.

657. Healy, W. B. Ingestion of soil by dairy cows. N. Z. J. Agric. Res. 11: 487-499, 1968.

658. Healy, W. B. In vitro studies on the effects of soil on elements in ruminal, "duodenal", and ileal liquors from sheep. N. Z. J. Agric. Res. 15:289-305, 1972.

659. Heathcote, J. G., and R. J. Washington. Analysis of the zinc-binding protein derived from the human benign hypertrophic prostate. J. Endocrinol. 58:421-423, 1973.

659a. Hedeskov, C. J., and K. Capito. The effect of starvation on insulin secretion and glucose metabolism in mouse pancreatic islets. Biochem. J. 140:423-433, 1974.

659b. Hegsted, D. M., J. M. McKibbin, and C. K. Drinker. The Biological, Hygienic, and Medical Properties of Zinc and Zinc Compounds. Supplement No. 179 to Public Health Reports. Washington, D. C.: U. S. Government Printing Office, 1945. 44 pp.

660. Hellwege, H. H., H. Schmalfuss, and D. Goschenhofer. Microchemische Zinkbestimmung im Serum und Urin. Z. Klin. Chem. Klin. Biochem. 7: 56-59, 1969.

660a. Hellwege, H. H. Tagesrhythmische Schwankungen des Serumzinkspiegels. Klin. Wochenschr. 48:1063-1064, 1970.

661. Helwig, H. L., E. M. Hoffer, W. C. Thielen, A. E. Alcocer, D. R. Hotelling, and W. H. Rogers. Modified zinc analysis method and serum and urinary zinc levels in control subjects. Amer. J. Clin. Path. 45: 160-165, 1966.

662. Hem, J. D. Chemistry and occurrence of cadmium and zinc in surface and groundwater. Water Resources Res. 8:661-679, 1972.

663. Hendricks, D. G., and A. W. Mahoney. Glucose tolerance in zinc-deficient rats. J. Nutr. 102:1079-1084, 1972.

664. Hendrickson, E. R. Air sampling and quantity measurement, pp. 3-13. In A. C. Stern, Ed. Air Pollution. Vol. 2. Analysis, Monitoring, and Surveying. (2nd ed.) New York: Academic Press, 1968.

665. Hendrickson, E. R. Sampling aerosol contaminants, pp. 24-34. In A. C. Stern, Ed. Air Pollution. Vol. 2. Analysis, Monitoring and Surveying. (2nd ed.) New York: Academic Press, 1968.

666. Henkin, R. I. Griseofulvin and dysgeusia: Implications? Ann. Intern. Med. 74:795-796, 1971.

667. Henkin, R. I. Growth-hormone-dependent changes in zinc and copper metabolism in man, pp. 652-655. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium on Trace Element Metabolism in Animals, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

668. Henkin, R. I. Metal-albumin-amino acid interactions: Chemical and physiological interrelationships. Adv. Exp. Med. Biol. 48:299-328, 1974.

669. Henkin, R. I. Newer aspects of copper and zinc metabolism, pp. 255-312. In W. Mertz and W. E. Cornatzer, Eds. Newer Trace Elements in Nutrition. New York: Marcel Dekker, Inc., 1971.

670. Henkin, R. I. On the role of adrenocorticosteroids in the control of zinc and copper metabolism, pp. 647-651. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium on Trace Element Metabolism in Animals held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

671. Henkin, R. I. Taste, pp. 468-483. In R. Hinchcliffe and D. Harrison, Eds. Scientific Foundations of Otolaryngology. London: William Heinemann Medical Books, Ltd., 1976.

672. Henkin, R. I. The definition of primary and accessory areas of olfaction as the basis for a classification of decreased olfactory acuity, pp. 235-252. In T. Hayashi, Ed. Olfaction and Taste II. Proceedings of the Second International Symposium held in Tokyo September 1965. New York: Pergamon Press, 1967.

672a. Henkin, R. I., and R. L. Aamodt. Zinc absorption in acrodermatitis enteropathica and in hypogeusia and hyposmia. Lancet 1:1379-1380, 1975.

672b. Delete--use 672a.

673. Henkin, R. I., H. R. Keiser, and D. Bronzert. Histidine-dependent zinc ion, hypogeusia, anorexia and hyposmia. J. Clin. Invest. 51:44a, 1972. (abstract)

673a. Henkin R. I., P. J. Schechter, W. T. Friedewald, D. L. DeMets, and M. Raff. A double blind study of the effects of zinc sulfate on taste and smell dysfunction. Amer. J. Med. Sci. 272:285-299, 1976.

674. Henkin, R. I., R. E. Lippoldt, J. Bilstad, and H. Edelhoch. A zinc protein isolated from human parotid saliva. Proc. Nat. Acad. Sci. U.S.A. 72:488-492, 1975.

675. Henkin, R. I., J. R. Marshall, S. Meret, and R. W. Bates. Trace metals in milk. Possible regulation by prolaction. Abstract 187, p. 130. in The Endocrine Society. Program of the 52nd Meeting, 1970. Bethesda, Maryland: Endocrine Society, 1970.

676. Henkin, R. I., J. R. Marshall, and S. Meret. Maternal-fetal metabolism of copper and zinc at term. Amer. J. Obstet. Gynecol. 110:131-134, 1971.

677. Henkin, R. I., S. Meret, and J. Jacobs. Steroid-dependent changes in copper and zinc metabolism. J. Clin. Invest. 48:38a, 1969. (abstract)

678. Henkin, R. I., C. W. Mueller, and R. O. Wolf. Estimation of zinc concentration of parotid saliva by flameless atomic absorption spectrophotometry in normal subjects and in patients with idiopathic hypogeusia. J. Lab. Clin. Med. 86:175-180, 1975.

679. Henkin, R. I., B. M. Patten, P. K. Re, and D. A. Bronzert. A syndrome of acute zinc loss. Cerebellar dysfunction, mental changes, anorexia, and taste and smell dysfunction. Arch. Neurol. 32:745-751, 1975.

680. Henkin, R. I., P. J. Schechter, R. C. Hoye, and C. F. T. Mattern. Idiopathic hypogeusia with dysgeusia, hyposmia and dysosmia. J.A.M.A. 217:434-440, 1971.

681. Henkin, R. I., P. J. Schechter, M. S. Raff, D. A. Bronzert, and W. T. Friedewald. Zinc and taste acuity: A clinical study including a laser microprobe analysis of the gustatory receptor area, pp. 204-228. In W. J. Pories, W. H. Strain, J. M. Hsu, and R. L. Woosley, Eds. Clinical Applications of Zinc Metabolism. Proceedings of the International Symposium. Springfield, Ill.: Charles C Thomas, 1974.

682. Henkin, R. I., J. D. Schulman, C. B. Schulman, and D. A. Bronzert. Changes in total, nondiffusible, and diffusible plasma zinc and copper during infancy. J. Pediatr. 82:831-837, 1973.

682a. Henkin, R. I., and F. R. Smith. Hyposmia in acute viral hepatitis. Lancet 1:823-826, 1971.

683. Henkin, R. I., and F. R. Smith. Zinc and copper metabolism in acute hepatitis. Amer. J. Med. Sci. 264:401-409, 1972.

684. Henkin, R. I., N. Talal, A. L. Larson, and C. F. T. Mattern. Abnormalities of taste and smell in Sjogren's syndrome. Ann. Intern. Med. 76:375-383, 1972.

684a. Henkin, R. I., and R. O. Wolf. On the role of parotid zinc, protein and the parotid zinc protein in taste. Clin. Res. 23:393A, 1975. (abstract)

685. Hennig, A., J. Martin, M. Anke, and D. Schüler. Die Parakeratose des Schweines. Arch. Exp. Veterinärmed. 23:911-920, 1969.

685a. Henzel, J. H., M. S. DeWeese, and E. L. Lichti. Zinc concentrations within healing wounds. Arch. Surg. 100:349-357, 1970.

686. Henzel, J. H., B. Holtmann, F. W. Keitzer, M. S. DeWeese, and E. Lichti. Trace elements in atherosclerosis, efficacy of zinc medication as a therapeutic modality, pp. 83-99. In D. D. Hemphill, Ed. Trace Substances in Environmental Health -- II. Proceedings of University of Missouri's 2nd Annual Conference, 1968. Columbia: University of Missouri, 1969.

686a. Henzel, J. H., F. W. Keitzer, E. L. Lichti, and M. S. DeWeese. Efficacy of zinc medication as a therapeutic modality in atherosclerosis: Followup observations on patients medicated over prolonged periods. pp. 336-341. In D. D. Hemphill, Ed. Trace Substances in Environmental Health -- IV. Proceedings of University of Missouri's 4th Annual Conference, 1970. Columbia: University of Missouri, 1971.

686b. Henzel, J. H., E. L. Lichti, W. Shepard, and J. Paone. Long-term oral zinc sulfate in the treatment of atherosclerotic peripheral vascular disease: Efficacy of possible mechanisms of action, pp. 243-259. In W. J. Pories, W. H. Strain, J. M. Hsu, and R. L. Woosley, Eds. Clinical Applications of Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1974.

687. Herbert, D. W. M., and D. S. Shurben. The toxicity to fish of mixtures of poisons. I. Salts of ammonia and zinc. Ann. Appl. Biol. 53: 33-41, 1964.

688. Herrick, J. B. The role of zinc in nutrition of food-producing animals (a review). Vet. Med. Small Anim. Clin. 69:85-89, 1974.

689. Herring, W. O. Secondary Zinc Industry Emission Control Problem Definition Study. Part 1 -- Technical Study. Final Report. Durham, N. C.: U. S. Environmental Protection Agency. Air Pollution Control Office, 1971. 150 pp.

690. Herzig, C. L., and C. C. Bigelow. The destabilization of bovine pancreatic ribonuclease. Biochem. Biophys. Res. Commun. 26:645-650, 1967.

691. Heth, D. A., W. M. Becker, and W. G. Hoekstra. Effect of calcium, phosphorous and zinc on zinc-65 absorption and turnover in rats fed semipurified diets. J. Nutr. 88:331-337, 1966.

692. Heth, D. A., and W. G. Hoekstra. Zinc-65 absorption and turnover in rats. I. A procedure to determine zinc-65 absorption and the antagonistic effect of calcium in a practical diet. J. Nutr. 85:367-374, 1965.

693. Hetland, Ø., and E. Brubakk. Diurnal variation in serum zinc concentration. Scand. J. Clin. Lab. Invest. 32:225-226, 1973.

694. Hewitt, E. J. Relation of manganese and some other metals to the iron status of plants. Nature 161:489-490. 1948.

695. Hewitt, E. J. Sand and Water Culture Methods Used in the Study of Plant Nutrition. Technical Communication No. 22. (Rev. 2nd ed.) Farnham Royal, England: Commonwealth Agricultural Bureaux, 1966. 547 pp.

696. Hewitt, W. B., and M. E. Gardner. Some studies of the absorption of zinc sulfate in Thompson seedless grape canes. Plant Physiol. 31:393-399, 1956.

697. Heyroth, F. F. Metal-fume fever, pp. 737-738. In F. A. Patty, Ed. Industrial Hygiene and Toxicology. Vol. 2. New York: Interscience Publishers, Inc., 1949.

698. Hiatt, A. J., and H. F. Massey. Zinc levels in relation to zinc content and growth of corn. Agron. J. 50:22-24, 1958.

699. Hibbard, P. L. A dithizone method for measurement of small amounts of zinc. Ind. Eng. Chem. Anal. Ed. 9:127-131, 1937.

700. Hibbard, P. L. Estimation of copper, zinc, and cobalt (with nickel) in soil extracts. Ind. Eng. Chem. Anal. Ed. 10:615-618, 1938.

701. Hilderbrand, D. C., and D. H. White. Trace-element analysis in hair: An evaluation. Clin. Chem. 20:148-151, 1974.

702. Hill, C. H. Influence of high levels of minerals on the susceptibility of chicks to Salmonella gallinarum. J. Nutr. 104:1221-1226, 1974.

703. Hill, C. H., G. Matrone, W. L. Payne, and C. W. Barber. In vivo interactions of cadmium with copper, zinc, and iron. J. Nutr. 80:227-235, 1963.

704. Hiltner, R. S., and H. J. Wichmann. Zinc in oysters. J. Biol. Chem. 38: 205-221, 1919.

705. Himes, F. L., and S. A. Barber. Chelating ability of soil organic matter. Soil. Sci. Soc. Amer. Proc. 21:368-373, 1957.

706. Himmelhoch, S. R. Leucine aminopeptidase: A zinc metalloenzyme. Arch. Biochem. Biophys. 134:597-602, 1969.

707. Hindriks, F. A., A. Groen, and A. M. Kroon. On the relation of heavy metals to the activity and heat stability of alkaline phosphatase from human placenta. Biochim. Biophys. Acta 315:94-102, 1973.

707a. Hinman, J. J., Jr. Desirable characteristics of a municipal water supply. J. Amer. Water Works Assoc. 30:484-494, 1938.

708. Hirose, M., E. Sugimoto, and H. Chiba. Studies on crystalline yeast phosphoglucomutase: The presence of intrinsic zinc. Biochim. Biophys. Acta 289:137-146, 1972.

709. Hoare, R., G. E. Delory, and D. W. Penner. Zinc and acid phosphatase in the human prostate. Cancer 9:721-726, 1956.

710. Hoch, F. L., and B. L. Vallee. Metabolic role of zinc, pp. 337-363. In C. A. Lamb, O. G. Bently, and J. M. Beattie, Eds. Trace Elements. Proceedings of the Conference held at the Ohio Agricultural Experiment Station, Wooster, Ohio, October 14-16, 1957. New York: Academic Press, 1958.

711. Hodgson, J. F. Chemistry of the micronutrient elements in soils. Adv. Agron. 15:119-159, 1963.

712. Hodgson, J. F., W. L. Lindsay, and W. D. Kemper. Contributions of fixed charge and mobile complexing agents to the diffusion of zinc. Soil Sci. Soc. Amer. Proc. 31:410-413, 1967.

713. Hodgson, J. F., W. L. Lindsay, and J. F. Trierweiler. Micronutrient cation complexing in soil solution: II. Complexing of zinc and copper in displaced solution from calcareous soils. Soil Sci. Soc. Amer. Proc. 30:723-726, 1969.

714. Deleted.

715. Hoekstra, W. G. Skeletal and skin lesions of zinc-deficient chickens and swine. Amer. J. Clin. Nutr. 22:1268-1277, 1969.

716. Hoekstra, W. G. The complexity of dietary factors affecting zinc nutrition and metabolism in chicks and swine, pp. 347-353. In C. F. Mills, Ed. Trace Element Metabolism in Animals. Proceedings of WAAP/IBP International Symposium, Aberdeen, Scotland, July 1969. London: E. & S. Livingston, 1970.

717. Höffken, B., and J.-G. Rausch-Stroomann. Die Zinkausscheidung bei Diabetikern unter Penicillamin. Z. Klin. Chem. Klin. Biochem. 7:4-7, 1969.

718. Hofman, H. O. [Word zinc first used in Europe], p. 3. In Metallurgy of Zinc and Cadmium. New York: McGraw-Hill Book Company, Inc., 1922.

719. Högl, O., and H. Sulser. Blei, Kupfer und Zink in Trink- und Brauchwasser. Mitteil. Gebiete Lebensmittel. Hyg. 42:286-311, 1951.

720. Hohn, C., and H. H. Evans. Identification of a low molecular weight ^{65}Zn complex in rat intestine. Proc. Soc. Exp. Biol. Med. 144: 793-795, 1973.

720a. Hohnadel, D. C., F. W. Sunderman, Jr., M. W. Nechay, and M. D. McNeely. Atomic absorption spectrometry of nickel, copper, zinc, and lead in sweat collected from healthy subjects during sauna bathing. Clin. Chem. 19:1288-1292, 1973.

721. Holtmeier, H. J., and M. Kuhn. Zink- und Magnesiummangel beim Menschen. Therapiewoche 22:4536-4546, 1972.

722. Hopkins, H. A., H. A. Campbell, B. Barbiroli, and V. R. Potter. Thymidine kinase and deoxyribonucleic acid metabolism in growing and regenerating livers from rats on controlled feeding schedules. Biochem. J. 136: 955-966, 1973.

723. Horčičko, J., J. Borovanský, and J. Duchon. Verteilung von Zink und Kupfer im menschlichen Kopfhaar verschiedener Farbtöne. Dermatol. Monatsschr. 159:206-209, 1973.

724. Horecker, B. L., O. Tsolas, and C. Y. Lai. Aldolases, pp. 213-258. In P. D. Boyer, Ed. The Enzymes. Vol. 7. Elimination and Addition. Aldol Cleavage and Condensation. Ohter C-C Cleavage. Phosphorolysis. Hydrolysis (Fats, Glycosides). (3rd ed.) New York: Academic Press, 1972.

725. Horwitz, Ed. Zinc--offical, final action, pp. 102-105. In Official Methods of Analysis of the Association of Offical Agricultural Chemists. (10th ed.) Washington, D. C.: Association of Offical Agricultural Chemists, 1965.

726. Hoss, D. E. Accumulation of zinc-65 by flounder of the genus Paralichthys. Trans. Amer. Fish. Soc. 93:364-368, 1964.

727. Hoss, D. E. Routine Energy Requirements of a Population of Pinfish (Lagodon rhomboides, Linnaeus) in the Newport River Estuary. Ph.D. Thesis. Raleigh: North Carolina State University, 1971. 87 pp.

728. Hsu, F. S., L. Krook, W. G. Pond, and R. Duncan. Lead, zinc and calcium interrelationships in growing pigs. J. Nutr. 103(7):xxiv, 1973. (abstract)

728a. Hsu, J. M. Zinc content in tissues of pyridoxine deficient rats. Proc. Soc. Exp. Biol. Med. 119:177-180, 1965.

729. Hsu, J. M. Hydroxyprolinuria in zinc deficient rats. Fed. Proc. 32:895, 1973. (abstract)

729a. Hsu, J. M., and W. L. Anthony. Effect of zinc deficiency and repletion on thymidine metabolism. Clin. Chem. 21:544-550, 1975.

730. Hsu, J. M., and W. L. Anthony. Effect of zinc deficiency on urinary excretion of nitrogenous compounds and liver amino acid-catabolizing enzymes in rats. J. Nutr. 105:26-31, 1975.

731. Hsu, J. M., and W. L. Anthony. Impairment of cystine-^{35}S incorporation into skin protein by zinc-deficient rats. J. Nutr. 101:445-452, 1970.

732. Hsu, J. M., and W. L. Anthony. Zinc deficiency and collagen synthesis in rat skin, pp. 137-143. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - VI. Proceedings of 6th Annual Conference on Trace Substances in Environmental Health held in Columbia, Missouri, June 13-15, 1972. Columbia: University of Missouri, 1973.

733. Hsu, J. M., and W. L. Anthony. Zinc deficiency and urinary excretion of taurine-^{35}S and inorganic sulfate-^{35}S following cystine-^{35}S injection in rats. J. Nutr. 100:1189-1196, 1970.

733a. Hsu, J. M., W. L. Anthony, and P. J. Buchanan. Zinc deficiency and oxidation of L-methionine-methyl-^{14}C in rats. J. Nutr. 97:279-285, 1969.

733b. Hsu, J. M., K. M. Kim, and W. L. Anthony. Biochemical and electron microscopic studies of rat skin during zinc deficiency. Adv. Exp. Biol. Med. 48:347-388, 1974.

734. Hsu, F. S., L. Krook, W. G. Pond, and J. R. Duncan. Interactions of dietary calcium with toxic levels of lead and zinc in pigs. J. Nutr. 105:112-118, 1975.

735. Hsu, J. M., and R. L. Woosley. Metabolism of L-methionine-^{35}S in zinc deficient rats. J. Nutr. 102:1181-1186, 1972.

736. Hsu, T. H. S., and J. M. Hsu. Zinc deficiency and epithelial wound repair An autoradiographic study of ^{3}H-thymidine incorporation. Proc. Soc. Exp. Biol. Med. 140:157-160, 1972.

737. Hsueh, A. M., M. Simonson, M. J. Kellum, and B. F. Chow. Perinatal undernutrition and the metabolic and behavioral development of the offspring. Nutr. Rep. Int. 7:437-445, 1973.

738. Hu, K. H., and R. L. Friede. Topographic determination of zinc in human brain by atomic absorption spectrophotometry. J. Neurochem. 15:677-685, 1968.

739. Huacuja, L., A. Sosa, N. M. Delgado, and A. Rosado. A kinetic study of the participation of zinc in human spermatozoa metabolism. Life Sci. 13:1383-1394, 1973.

740. Huber, A. M., and S. N. Gershoff. Effect of zinc deficiency in rats on insulin release from the pancreas. J. Nutr. 103:1739-1744, 1973.

741. Huber, A. M., and S. N. Gershoff. Effects of dietary zinc and calcium on the retention and distribution of zinc in rats fed semipurified diets. J. Nutr. 100:949-954, 1970.

742. Huber, A. M., and S. N. Gershoff. Effects of dietary zinc on zinc enzymes in the rat. J. Nutr. 103:1175-1181, 1973.

743. Huber, A. M., and S. N. Gershoff. In vitro effects of zinc on insulin activity in adipose tissue. Proc. Soc. Exp. Biol. Med. 123:352-356, 1966.

743a. Hübscher, J., N. Presselt, K.-J. Halbhuber and G. Geyer. Autoradiographische Untersuchungen über die Verteilung einiger Metallisotope in Organen von Wistar-Ratten. Z. Mikrosk. Anat. Forsch. 86:553-559, 1972.

744. Huffman, E. W. D., Jr., and J. F. Hodgson. Distribution of cadmium and zinc/cadmium ratios in crops from 19 states east of the Rocky Mountains. J. Environ. Qual. 2:289-291, 1973.

745. Huggett, R. J., M. E. Bender, and H. D. Slone. Utilizing metal concentration relationships in the eastern oyster (Crassostrea virginica) to detect heavy metal pollution. Water Res. 7:451-460, 1973.

746. Huggett, R. J., F. A. Cross, and M. E. Bender. Distribution of copper and zinc in oysters and sediments from three coastal-plain estuaries, pp. 224-238. In F. G. Howell, J. B. Gentry and M. H. Smith, Eds. Mineral Cycling in Southeastern Ecosystems. Proceedings of a Symposium, 1974. CONF-740513. Oak Ridge, Tenn.: U. S. Energy Research and Development Administation, 1975.

746a. Hughes, B. O., and W. A. Dewar. A specific appetite for zinc in zinc-depleted domestic fowls. Brit. Poult. Sci. 12:255-258, 1971.

747. Hulett-Cowling, F. M., and L. L. Campbell. Molecular weight and subunits of the alkaline phosphatase of Bacillus licheniformis. Biochemistry 10:1371-1376, 1971.

748. Hulett-Cowling, F. M., and L. L. Campbell. Purification and properties of an alkaline phosphatase of Bacillus licheniformis. Biochemistry 10:1364-1371, 1971.

749. Hull, R. L. Use of trace elements in intravenous hyperalimentation solutions. Amer. J. Hosp. Pharmacol. 31:759-761, 1974.

750. Hunt, C. E., and W. W. Carlton. Cardiovascular lesions associated with experimental copper deficiency in the rabbit. J. Nutr. 87:385-393, 1965.

751. Hunter, D. Metal fume fever, pp. 421-423. In The Diseases of Occupation. (4th ed.) Boston: Little Brown and Co., 1969.

752. Hunter, F. E., Jr., and L. Ford. Inactivation of oxidative and phosphorylative systems in mitochondria by preincubation with phosphate and other ions. J. Biol. Chem. 216:357-369, 1955.

753. Hunter, J. G., and O. Vergnano. Trace-element toxicities in oat plants. Ann. Appl. Biol. 40:761-777, 1953.

754. Hupka, E. Über Flugstaubvergiftungen in der Umgebung von Metallhütten. Wien. Tierärztl. Monatsschr. 42:763-775, 1955.

754a. Hurley, L. S. Zinc deficiency, potatoes, and congenital malformations in man. Teratology 10:205-206, 1974. (letter)

755. Hurley, L. S., J. Gowan, and G. Milhaud. Calcium metabolism in manganese-deficient and zinc-deficient rats. Proc. Soc. Exp. Biol. Med. 130: 856-860, 1969.

756. Hurley, L. S., J. Gowan, and H. Swenerton. Teratogenic effects of short-term and transitory zinc deficiency in rats. Teratology 4:199-204, 1971.

757. Hurley, L. S., and P. B. Mutch. Prenatal and postnatal development after transitory gestational zinc deficiency in rats. J. Nutr. 103:649-655, 1973.

758. Hurley, L. S., and R. E. Shrader. Abnormal development of preimplantation rat eggs after three days of maternal dietary zinc deficiency. Nature 254:427-429, 1975.

759. Hurley, L. S., and R. E. Shrader. Congenital malformations of the nervous system in zinc-deficient rats. Int. Rev. Neurobiol. Suppl. 1: 7-51, 1972.

760. Hurley, L. S., K. Sucher, D. Story, and G. Cosens. Interaction of dietary protein and zinc in the pregnant rat. J. Nutr. 103(7):xxv, 1973. (abstract)

761. Hurley, L. S., and H. Swenerton. Lack of mobilization of bone and liver zinc under teratogenic conditions of zinc deficiency in rats. J. Nutr. 101:597-604, 1971.

761a. Hurley, L. S., and H. Swenerton. Congenital malformations resulting from zinc deficiency in rats. Proc. Soc. Exp. Biol. Med. 123:692-696, 1966.

762. Hurley, L. S., and S.-H. Tao. Alleviation of teratogenic effects of zinc deficiency by simultaneous lack of calcium. Amer. J. Physiol. 222:322-325, 1972.

762a. Husain, S. L. Oral zinc sulphate in leg ulcers. Lancet 1:1069-1071, 1969.

762b. Hutchinson, D. W., B. Johnson, and A. J. Knell. Metal complexes of bilirubin in aprotic solvents. Biochem. J. 133:399-400, 1973.

763. Hutchinson, T. C. The Effect of Traffic Density on Levels of Lead in Soils and Vegetation of Metro Toronto. Paper Presented at INTECOL (International Association for Ecology) Symposium on Physiological Ecology of Plants and Animals in Extreme Environments, Dubrovnik, Yugoslavia, 1972.

764. Hutchinson, T. C., and H. Czyrska. Cadmium and zinc toxicity and synergism to floating aquatic plants, pp. 59-65. In Water Pollution Research in Canada, 1972. Proceedings of the Seventh Canadian Symposium on Water Pollution Research, 1972.

765. Hutchinson, T. C., and L. M. Whitby. A study of the airborne contamination of vegetation and soils by heavy metals from the Sudbury, Ontario, copper-nickel smelters, pp. 179-189. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - VII. Proceedings of 7th Annual Conference held in Columbia, Missouri, June 12-14, 1973. Columbia: University of Missouri, 1973.

766. Hwang, J. Y. Atomic absorption and flame emission spectroscopy in the analysis of trace metals in atmospheric particulates, pp. 352-356. In H. M. Englund and W. T. Beery, Eds. Proceedings of the Second International Clean Air Congress. Held at Washington, D. C., Dec. 6-11, 1970. New York: Academic Press, 1971.

767. Hwang, J. Y. Trace metals in atmospheric particulates and atomic-absorption spectroscopy. Anal. Chem. 44(14):20A-27A, 1972.

767a. Pike, R. L., and M. L. Brown. The components of protein and fat storage in pregnancy, p. 333. In Nutrition: An Integrated Approach. New York: John Wiley & Sons, Inc., 1967.

768. Ikuta, K. Studies on accumulation of heavy metals in aquatic organisms. IV. On disappearance of abnormally accumulated copper and zinc in oysters. Bull. Jap. Soc. Sci. Fish. 34:482-487, 1968. (in Japanese, summary in English)

769. Im, M. J. C., J. M. Hsu, and J. E. Hoopes. Enzyme activities in the epidermis of zinc-deficient rats. Fed. Proc. 34:906, 1975. (abstract)

770. Imai, S., K. Ito, A. Hamaguchi, Y. Kusaka, and M. Warashina. Emission spectrographic determination of trace elements in airborne particulates using membrane filter. Bunseki Kagaku (Jap. Anal.) 22:551-558, 1973. (in Japanese, summary in English)

771. International Labor Office/World Health Organization Committee on Occupational Health. Permissible Levels of Toxic Substances in the Working Environment. Sixth Session of the Joint ILO/WHO Committee on Occupational Health, Geneva 4-10, June 1968. Geneva: International Labour Office, 1970. 405 pp.

772. Intersociety Committee. Methods of Air Sampling and Analysis. Washington, D. C.: American Public Health Association, 1972. 480 pp.

773. Intersociety Committee. Tentative method of analysis for dustfall from the atmosphere, pp. 373-375. In Methods of Air Sampling and Analysis. Washington, D. C.: American Public Health Association, 1972.

774. Intersociety Committee. Tentative method of analysis for suspended particulate matter in the atmosphere: (high volume method), pp. 365-372. In Methods of Air Sampling and Analysis. Washington, D. C.: American Public Health Association, 1972.

775. Iqbal, M. Activity of alkaline phosphatase and carbonic anhydrase in male and female zinc-deficient rats. Enzyme 12:33-40, 1971.

776. Isaac, R. A., and J. D. Kerber. Atomic absorption and flame photometry: Techniques and uses in soil, plant, and water analysis, pp. 17-37. In L. M. Walsh, Ed. Instrumental Methods for Analysis of Soils and Plant Tissue. Madison, Wis.: Soil Science Society of America, 1971.

777. Delete 777--use 769.

778. Delete 778--use 770

779. Delete 779--use 771

780. Delete 780--use 772

781. Delete 781--use 773

782. Delete 782--use 774

783. Delete 783--use 775

783a. Iqbal, M. Effect of in vitro addition of zinc on alkaline phosphatase activity in the zinc-deficient rat. Enzyme Biol. Clin. 11:412-422, 1970.

784. Delete 784--use 776

785. Isaacson, A., and A. Sandrow. Effects of zinc on responses of skeletal muscle. J. Gen. Physiol. 46:655-677, 1963.

786. Jackson, T. L., J. Hay, and D. P. Moore. The effect of Zn on yield and chemical composition of sweet corn in the Willamette Valley. Proc. Amer. Soc. Hort. Sci. 91:462-471, 1967.

787. Jacob, R. A., L. M. Klevay, and E. J. Thacker. Hypercholesterolemia due to meat anemia. Fed. Proc. 34:899, 1975. (abstract)

788. Jacobs, F. A., T. W. Winter, and H. H. Sandstead. Lymph proteins in zinc deficient rats. Fed. Proc. 34:922, 1975. (abstract)

788a. Jaffe, N. R., and E. M. Johnson. Alterations in the ontogeny and specific activity of phosphomonoesterases associated with abnormal chondrogenesis and osteogenesis in limbs of fetuses from folic-acid deficient pregnant rats. Teratology 8:33-49, 1973.

789. Jagannathan, V., K. Singh, and M. Damodaran. Carbohydrate metabolism in citric acid fermentation. 4. Purification and properties of aldolase from Aspergillus niger. Biochem. J. 63:94-105, 1956.

789a. Janick, J., L. Zeitz, and W. F. Whitmore, Jr. Seminal fluid and spermatozoon zinc levels and their relationship to human spermatozoon motility. Fertil. Steril. 22:573-580, 1971.

789b. Japan Association of Industrial Health. Recommendation for permissible concentrations, etc. (1971). Sangyo Igaku (Jap. J. Ind. Health) 13:475-484, 1971. (in Japanese)

790. Javillier, M. Influence du zinc sur la consommation par l'Aspergillus niger des ses aliments hydrocarbonés, azotés et minéraux. C. R. Acad. Sci. D (Paris) 155:190-193, 1912.

790a. Jelínek, J. M., O. Marhan, and M. Šeda. The effect of copper sulphate on pituitary LH in rats. Endocrinol. Exp. (Bratisl.) 4:37-43, 1970.

791. Jenne, E. A. Controls on Mn, Fe, Co, Ni, Cu, and Zn concentrations in soils and water: The significant role of hydrous Mn and Fe oxides, pp. 337-387. In R. F. Gould, Ed. Trace Inorganics in Water. Advances in Chemistry Series 73. Washington, D. C.: American Chemical Society, 1968.

792. Jennings, C. D., and C. Osterberg. Sediment radioactivity in the Columbia River estuary, pp. 300-306. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. RID-4500. Oak Ridge, Tenn: U. S. Atomic Energy Commission, 1969.

793. Jensen, H. L., and C. G. Lamm. On the zinc content of Danish soils. Acta Agric. Scand. 11:63-81, 1961.

794. John, M. K. Influence of soil properties and extractable zinc on zinc availability. Soil Sci. 113:222-227, 1972.

795. John, W., R. Kaifer, K. Rahn, and J. J. Wesolowski. Trace element concentrations in aerosols from the San Francisco Bay Area. Atmos. Environ. 7:107-118, 1973.

796. Johnson, D., Jr., A. L. Mehring, Jr., F. X. Savino, and H. W. Titus. The tolerance of growing chickens for dietary zinc. Poult. Sci. 41:311-317, 1962.

797. Johnson, N. C. Study of copper and zinc metabolism during pregnancy. Proc. Soc. Exp. Biol. Med. 108:518-519, 1961.

798. Johnson, W. J., and W. G. Schrenk. Nature of zinc-containing substances in the alfalfa plant cell. J. Agric. Food Chem. 12:210-213, 1964.

799. Jones, J. B., Jr. Distribution of fifteen elements in corn leaves. Commun. Soil Sci. Plant Anal. 1:27-34, 1970.

800. Jones, J. B., Jr. Plant tissue analysis for micronutrients, pp. 319-346. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

800a. Janes, J. M., J. T. McCall, and L. R. Elveback. Trace metals in human osteogenic sarcoma. Mayo Clin. Proc. 47:476-478, 1972.

801. Judy, W., G. Lessman, T. Rozycka, L. Robertson, and B. Ellis. Field and laboratory studies with zinc fertilization of pea beans. Mich. Agric. Exp. Stat. Q. Bull. 46:386-400, 1964.

802. Judy, W. J. Melton, G. Lessman, B. Ellis, and J. Davis. Field and Laboratory Studies with Zinc Fertilization of Pea Beans, Corn and Sugar Beets in 1964. Agricultural Experiment Station Research Report 33. East Lansing: Michigan State University, 1965. 8 pp.

802a. Juljulian, H. H., and A. K. Kurban. Acantholysis: A feature of acrodermatitis enteropathica. Arch. Derm. 103:105-106, 1971.

803. Jyung, W. H., M. E. Camp, D. E. Polson, M. W. Adams, and S. H. Wittwer. Differential response of two bean varieties to zinc as revealed by electrophoretic pattern. Crop Sci. 12:26-29, 1972.

804. Jyung, W. H., A. Ehmann, K. Schlender, and J. Scala. Zinc nutrition and starch metabolism in Phaseolus vulgaris L. Plant Physiol. 55:414-420, 1975.

805. Jyung, W. H., and K. K. Schlender. Zinc metabolism in higher plants. Its relation to starch synthetase. Plant Physiol. 47(Suppl.):6, 1971. (abstract)

806. Kägi, J. H. R., S. R. Himmelhoch, P. D. Whanger, J. L. Bethune, and B. L. Vallee. Equine hepatic and renal metallothioneins. Purification, molecular weight, amino acid composition, and metal content. J. Biol. Chem. 249:3537-3542, 1974.

807. Kägi, J. H. R., and B. L. Vallee. Metallothionein: A cadmium- and zinc-containing protein from equine renal cortex. J. Biol. Chem. 235:3460-3465, 1960.

808. Kägi, J. H. R., and B. L. Vallee. The role of zinc in alcoholic dehydrogenase. V. The effect of metal-binding agents on the structure of the yeast alcohol dehydrogenase molecule. J. Biol. Chem. 235:3188-3192, 1960.

809. Kahn, A. M., H. L. Helwig, A. G. Redeker, and T. B. Reynolds. Urine and serum zinc abnormalites in diseases of the liver. Amer. J. Clin. Path. 44:426-435, 1965.

810. Kahn, A. M., and R. S. Ozeran. Liver and serum zinc abnormalities in rats with cirrhosis. Gastroenterology 53:193-197, 1967.

811. Kahn, H. L., and J. S. Sebestyen. The determination of lead in blood and urine by atomic absorption spectrophotometry, with the sampling boat system. Atom. Absorpt. Newslett. 9:33-38, 1970.

812. Kaindl, F., P. Kühn, P. Holzhey, and M. Niederberger. Herztherapie mit Zink-Protamin-Glucagon. Verh. Dtsch. Ges. Inn. Med. 78:1099-1101, 1972.

813. Kaltenbach, T., and E. Eger. Beiträge zum histochemischen Nachweis von Eisen, Kupfer und Zink in der menschlichen Leber unter besonderer Berücksichtigung des Silbersulfid-Verfahrens nach Timm. Acta Histochem. 25:329-354, 1966.

814. Kampa, E. M. The euphausiid eye-- a re-evaluation. Vision Res. 5:475-481, 1965.

814a. Kampschmidt, R. F., and H. F. Upchurch. Effect of leukocytic endogenous mediator on plasma fibrinogen and haptoglobin. Proc. Soc. Exp. Biol. Med. 146:904-907, 1974.

814b. Kampschmidt, R. F., and H. F. Upchurch. The effect of endogenous pyrogen on the plasma zinc concentration of the rat. Proc. Soc. Exp. Biol. Med. 134:1150-1152, 1970.

815. Kane, P. F., and G. B. Larrabee. Trace analysis techniques for solids, pp. 33-68. Ann. Rev. Materials Sci. 2:33-68, 1972.

816. Kapur, S. P., B. R. Bhussry, S. Rao, and E. Harmuth-Hoene. Percutaneous uptake of zinc in rabbit skin. Proc. Soc. Exp. Biol. Med. 145: 932-937, 1974.

817. Kar, A. B., R. P. Das, and B. Mukerji. Prevention of cadmium induced changes in the gonads of rat by zinc and selenium -- a study in antagonism between metals in the biological system. Proc. Nat. Inst. Sci. India Part B 26(Suppl):40-50, 1960.

818. Kar, A. B., and V. P. Kamboj. Cadmium damage of the rat testis & its prevention. Indian J. Exp. Biol. 3:45-49, 1965.

819. Karlinsky, V. M., and P. A. Roomere. Changes in the zinc content in the blood serum and urine in infectious hepatitis. Klin. Med. 43(2): 78-83, 1965. (in Russian, summary in English)

820. Karvánek, M., and J. Böhmová. The content of copper, iron, nickel, manganese, zinc and molybdenum in spinach leaves. Sb. Vys. Skoly Chem. Technol. Prague E11:73-82, 1966.

821. Kasarskis, E. J., and W. G. Hoekstra. Effect of alterations of zinc status on the zinc content of the gastrointestinal tract of chicks. Proc. Soc. Exp. Biol. Med. 145:508-512, 1974.

822. Kasperek, K., H. Schicha, A. Höck, V. Siller, and L. E. Feinendegen. Serum-zink in Abhängigkeit von der Tageszeit und Nahrungsaufnahme. Strahlentherapie 145:229-233, 1973.

823. Katz, M. Problems in analysis of air contaminants, pp. 124-129. In B. Westley, Ed. Proceedings of the International Symposium on Identification and Measurement of Environmental Pollutants. Ottawa, Ontario, Canada, June 14-17, 1971. Ottawa: National Research Council of Canada, 1971.

824. Kawabata, T. Studies on the radiological contamination of fishes. III. Radiochemical studies of contaminated fish. Jap. J. Med. Sci. Biol. 8:359-372, 1955.

825. Kawashima, R., S. W. Kim, and S. Uesaka. Studies on importance of trace elements in farm animal feeding. XXXVIII. Effects of amino acid and protein on trace element toxicity in cellulose digestion by rumen bacteria. Bull. Res. Inst. Food Sci., Kyoto Univ. 32:8-16, 1969. (in Japanese, summary in English)

826. Kečkeš, S., B. Ozretić, and M. Krajnović. Loss of Zn^{65} in the mussel Mytilus galloprovincialis. Malacologia 7:1-6, 1968.

827. Keefer, R. F., R. N. Singh, D. J. Horvath, and P. R. Henderlong. Response of corn to time and rate of phosphorus and zinc application. Soil Sci. Soc. Amer. Proc. 36:628-632, 1972.

828. Keele, B. B., Jr., J. M. McCord, and I. Fridovich. Superoxide dismutase from Escherichia coli B. A new manganese-containing enzyme. J. Biol. Chem. 245:6176-6181, 1970.

829. Kehoe, R. A. Metal fume fever. Amer. Ind. Hyg. Assoc. Q. 9:66-70, 1948.

830. Keilin, D., and T. Mann. Activity of purified carbonic anhydrase. Nature 153:107-108, 1944. (letter)

831. Keilin, D., and T. Mann. Carbonic anhydrase. Purification and nature of the enzyme. Biochem. J. 34:1163-1176, 1940.

832. Keleti, T. Studies on D-glyceraldehyde-3-phosphate dehydrogenases. XXI. Some data on the binding of Zn in the enzyme. Biochim. Biophys. Acta 89:422-430, 1964.

833. Keleti, T. The role of zinc ions, -SH groups, and histidyl residues in the mechanism of dehydrogenases, pp. 103-120. In H. Sund, Ed. Pyridine Nucleotide-Dependent Dehydrogenases. Proceedings of an Advanced Study Institute held at the University of Konstanz, Germany, September 15-20, 1969. New York: Springer-Verlag, 1970.

834. Keleti, T. Zn in yeast D-glyceraldehyde-3-phosphate dehydrogenase. Biochem. Biophys. Res. Commun. 22:640-643, 1966.

835. Kelly, J. J. Neutron-activation analysis, pp. 535-556. In M. Zief and R. Speights, Eds. Ultrapurity: Methods and Techniques. New York: Marcel Dekker, Inc., 1972.

835a. Kerr, W. K., A. G. Keresteci, and H. Mayoh. The distribution of zinc within the human prostate. Cancer 13:550-554, 1960.

836. Kershaw, J. B. C. The Recovery and Use of Industrial and Other Wastes. London: Ernest Benn Ltd., 1928. 211 pp.

837. Kessler, B. Ribonuclease as a guide for the determination of zinc deficiency in orchard trees, pp. 314-322. In W. Reuther, Ed. Plant Analysis and Fertilizer Problems. Washington, D. C.: American Institute of Biological Science, 1961.

838. Kessler, B., and S. P. Monselise. Studies on ribonuclease, ribonucleic acid and protein synthesis in healthy and zinc-deficient citrus leaves. Physiol. Plant. 12:1-7, 1959.

839. Ketcheson, M. R., G. P. Barron, and D. H. Cox. Relationship of maternal dietary zinc during gestation and lactation to development and zinc, iron and copper content of the postnatal rat. J. Nutr. 98:303-311, 1969.

840. Kimura, K., and J. Kumura. Preliminary reports on the metabolism of trace elements in neuro-psychiatric diseases. I. Zinc in schizophrenia. Proc. Jap. Acad. 41:943-947, 1965.

841. King, L. D., and H. D. Morris. Land disposal of liquid sewage sludge: I. The effect on yeild, in vivo digestibility, and chemical composition of coastal bermudagrass (Cynodon dactylon L. Pers.). J. Environ. Qual. 1:325-329, 1972.

842. King, L. D., and H. D. Morris. Land disposal of liquid sewage sludge: II. The effect on soil pH, manganese, zinc and growth and chemical composition of rye (Secale cereale L.). J. Environ. Qual. 1:425-429, 1972.

843. Kinnamon, K. E. Copper, molybdenum and zinc interrelationships; the influence of inorganic sulfate upon distribution and excretion of ^{65}Zn and ^{90}Mo in pregnant rats. J. Nutr. 89:365-372, 1966.

843a. Kinnamon, K. E. Radiation and wound healing: Influence of dietary methionine and zinc on zinc-65 distribution and excretion in the rat. Radiat. Res. 29:184-193, 1966.

844. Kinnamon, K. E., and G. E. Bunce. Effects of copper, molybdenum, and zinc on zinc-65 tissue distribution and excretion in the rat. J. Nutr. 86:225-230, 1965.

845. Kirchgessner, M. Availability of some trace elements. Proc. Nutr. Soc. 24:89-99, 1965.

846. Kirchgessner, M., and J. Pallauf. Zinkrepletion in Serum und Leber wachsender Ratten. 4. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 29:77-85, 1972.

847. Kirchgessner, M., and J. Pallauf. Zinkdepletion wachsender Ratten. Leber, Knochen, Schwanz und Ganskörper. 3. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 29:65-76, 1972.

848. Kirchgessner, M., and J. Pallauf. Zum Einfluss von Fe-, Co- bzw. Ni-Zulagen bei Zinkmangel. Z. Tierphysiol. 31:268-274, 1973.

849. Kirchgessner, M., and H.-P. Roth. Beziehungen zwischen klinischen Mangelsymptomen und Enzymaktivitäten bei Zinkmangel. Zentralbl. Veterinaermed. (A) 22:14-26, 1975.

850. Kirchgessner, M., F. J. Schwarz, and E. Grassmann. Intestinal absorption of copper and zinc after dietary depletion. Bioinorg. Chem. 2:255-262, 1973.

851. Klein, D. H. Mercury and other metals in urban soils. Environ. Sci. Technol. 6:560-562, 1972.

851a. Klein, S. Störungen der Sofortadaptation und der Blendungsempfindlichkeit bei Patienten mit chronischen Lebererkrankungen und ihre Bedeutung für die Kraftfahrtauglichkeit. Dtsch. Gesundheitswesen 27:1235-1238, 1972.

852. Kleinman, L. J., H. G. Petering, and J. M. Sutherland. Blood carbonic anhydrase activity and zinc concentration in infants with respiratory-distress syndrome. New Engl. J. Med. 277:1157-1161, 1967.

853. Kleinman, M. T., T. J. Kneip, and M. Eisenbud. Meteorological influences on airborne trace metals and suspended particulates, pp. 161-166. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - VII. Proceedings of 7th Annual Conference held in Columbia, Missouri, June 12-14, 1973. Columbia: University of Missouri, 1973.

854. Klevay, L. M. An association between the amount of fat and the ratio of zinc to copper in 71 foods. Inferences about the epidemiology of coronary heart disease. Nutr. Rep. Int. 9:393-399, 1974.

854a. Klevay, L. M. Interactions among dietary copper, zinc, and the metabolism of cholesteral and phospholipids, pp. 553-556. In H. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals--2. Proceedings of the 2nd International Symposium, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

855. Klevay, L. M. Hair as a biopsy material. Amer. J. Clin. Nutr. 23:284-289, 1970.

856. Klevay, L. M. Hair as a biopsy material. IV. Geographic variations in the concentration of zinc. Nutr. Rep. Int. 10:181-187, 1974.

857. Klevay, L. M. Hypercholesterolemia in rats induced by ascorbic acid. Fed. Proc. 34:899, 1975. (abstract)

858. Klevay, L. M. Hypercholesterolemia in rats produced by an increase in the ratio of zinc to copper ingested. Amer. J. Clin. Nutr. 26:1060-1068, 1973.

858a. Klevay, L. M. The ratio of zinc to copper of diets in the United States. Nutr. Rep. Int. 11:237-242, 1975.

858b. Klevay, L. M., G. W. Evans, and H. H. Sandstead. Zinc/copper hypercholesterolemia: The effect of sodium phytate. Clin. Res. 23: 460A, 1975. (abstract)

859. Deleted.

859a. Knauer, G. A., and J. H. Martin. Seasonal variations of cadmium, copper, manganese, lead and zinc in water and phytoplankton in Monterey Bay, California. Limnol. Oceanogr. 18:597-604, 1973.

860. Knauss, H. J., and J. W. Porter. The absorption of inorganic ions by Chlorella pyrenoidosa. Plant Physiol. 29:229-234, 1954.

860a. Kneip, T. J., and M. Eisenbud. Trace Metals in Urban Aerosols. First Annual Progress Report. New York: New York University Medical Center, Institute of Environmental Medicine, 1973.

861. Kneip, T. J., M. Eisenbud, C. D. Strehlow, and P. C. Freudenthal. Airborne particulates in New York City. J. Air Pollut. Control Assoc. 20:144-149, 1970.

862. Kobayashi, J. Air and water pollution by cadmium, lead, and zinc attributed to the largest zinc refinery in Japan, pp. 117-128. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - V. Proceedings of 5th Annual Conference on Trace Substances in Environmental Health held in Columbia, Missouri, June 29 - July 1, 1971. Columbia: University of Missouri, 1972.

863. Kobayashi, J. Relation between the "itai-itai" disease and the pollution of river water with cadmium from a mine. Adv. Water Pollut. Res. 1: I-25/1--I25/7, 1971.

863a. Koch, H. J., Jr., E. R. Smith, and J. McNeely. Analysis of trace elements in human tissues. II. The lymphomatous disease. Cancer 10:151-160, 1957.

863b. Kobayashi, J. On geographical relationship beweeen the chemical nature of river water and death-rate from apoplexy. (Preliminary report). Ber. Ohara Inst. Landwirtschaftl. Biol. 11:12-21, 1957.

864. Kocsis, J. J., E. J. Walaszek, E. E. Graham, and E. M. K. Geiling. Zinc content of various pituitary fractions. Fed. Proc. 12:336-337, 1953.

865. Kohrs, M. B., J. Sauvage, R. E. Shank, R. Brennan, H. Siegel, and J. Nordstrom. Nutritional status of pregnant black women attending a prenatal clinic. Fed. Proc. 34:896, 1975. (abstract)

865a. Kollmer, W. E., P. Schramel, and K. Samsahl. Simultaneous determination of nine elements in some tissues of the rat using neutron activation analysis. Phys. Med. Biol. 17:555-562, 1972.

866. Kometani, T. Y., J. L. Bove, B. Nathanson, S. Siebenberg, and M. Magyar. Dry ashing of airborne particulate matter on paper and glass fiber filters for trace metal analysis by atomic absorption spectrometry. Environ. Sci. Technol. 6:617-620, 1972.

866a. Delete 866a -- use 871b.

866b. Kopito, L. E., H. J. Kosasky, S. H. Sturgis, B. L. Lieberman, and H. Shwachman. Water and electrolytes in human cervical mucus. Fertil. Steril. 24:499-506, 1973.

867. Kopito, L. E., H. Shwachman, G. F. Vawter, and J. Edlow. The pancreas in cystic fibrosis: Chemical composition and comparative morphology. Pediatr. Res. 10:742-749, 1976.

867a. Kopito, L. E., and H. Shwachman. Alterations in the elemental composition of hair in some diseases, pp. 83-90. In A. C. Brown, Ed. The First Human Hair Symposium. New York: Medcom Press, 1974.

867b. Delete 867b.-- use 151.

867c. Kopito, L., and H. Shwachman. Mineral composition of meconium. J. Pediatr. 68:313-314, 1966.

868. Kopito, L., and H. Shwachman. Spectroscopic analyses of tissues from patients with cystic fibrosis and controls. Nature 202:501-502, 1964.

869. Kopp, J. F., and R. C. Kroner. Trace Metals in Waters of the United States. A Five-year Summary of Trace Metals in Rivers and Lakes of the United States (Oct. 1, 1962-Sept. 30, 1967). Cincinnati: U. S. Department of the Interior. Federal Water Pollution Control Administration, Division of Pollution Surveillance, 1969. 205 pp.

870. Delete 870--use 871

871. Korant, B. D., J. C. Kauer, and B. E. Butterworth. Zinc ions inhibit replication of rhinoviruses. Nature 248:588-590, 1974.

871a. Kosasky, H. J., L. E. Kopito, S. H. Sturgis, and H. Shwachman. Changes in water and electrolytes in human cervial mucus during treatment with chlormadinone acetate. Fertil. Steril. 24:507-511, 1973.

872. Kowal, J., T. Cremona, and B. L. Horecker. Fructose 1, 6-diphosphase aldolase of Candida utilis: Purification and properties. Arch. Biochem. Biophys. 114:13-23, 1966.

873. Kowarski, S., C. S. Blair-Stanek, and D. Schachter. Active transport of zinc and identification of zinc-binding protein in rat jejunal mucosa. Amer. J. Physiol. 226:401-407, 1974.

873a. Delete -- use 863b.

874. Kozlowski, J., J. Litwin, and S. Sitniewski. A histochemical study of the granulocyte zinc content in cutaneous neoplasia and in skin reactions to internal malignancy. Acta Derm. Venereol. 47:269-274, 1967.

875. Kratzer, F. H., J. B. Allred, B. J. Davis, B. J. Marshall, and P. Vohra. The effect of autoclaving soybean protein and the addition of ethylenediaminetetraacetic acid on the biological availability of dietary zinc for turkey poults. J. Nutr. 68:313-322, 1959.

876. Krause, L. A. Aspects of Metal Fume Fever. D.S.I.H. Dissertation. University of Cincinnati, 1962. 95 pp.

877. Krishna Murthy, A. S., G. F. Vawter, L. Kopito, and E. Rosen. Retinal atrophy and cataract in rats following administration of N-methyl-N-nitrosourea. Proc. Soc. Exp. Biol. Med. 139:84-87, 1972.

878. Kristiansen, P. H. Zinc metabolism in swine. V. Influence of zinc deficiency on protein synthesis. Aarsberetn. Inst. Sterilitetsforsk. Kgl. Vet. Landbohejsk. 16:75-80, 1973. (in Danish, summary in English)

879. Kristiansen, P. H., and I. H. Pedersen. Zinc metabolism in swine. II. Variations in the content of zinc in blood during the early postnatal life. Aarsberetn. Inst. Sterilitetsforsk. Kgl. Vet. Landbohoejsk. 15:9-26, 1972. (in Danish, summary in English)

880. Kristiansen, P. H., I. H. Pedersen, and I. Wegger. Zinc metabolism in swine. I. Assessment of zinc status in pigs by analysis of blood, hair, and liver. Aarsberetn. Inst. Sterilitetsforsk. Kgl. Vet. Landbohoejsk. 14: 111-126, 1971. (in Danish, summary in English)

881. Kristiansen, P. H., and I. Wegger. Zinc metabolism in swine. VI. Absorption of ^{65}Zn from duodenum and *in vitro* uptake by different segments of the small intestine. Aarsberetn. Inst. Sterilitetsforsk. Kgl. Vet. Landbohoejsk. 17:33-46, 1974. (in Danish)

881a. Kroneman, J., G. J. W. v.d. Mey, and A. Helder. Hereditary zinc deficiency in Dutch Friesian cattle. Zentralbl. Veterinaermed. A 22:201-208, 1975.

882. Krylova, N. A. Determination of zinc in the air. Gig. Sanit. 34(10): 65-66, 1969. (in Russian)

883. Kujala, N. F., I. L. Larsen, and C. L. Osterberg. Radioisotope measurements of the viscera of Pacific salmon, pp. 440-449. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. Tid-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

884. Kumar, K. S. V., K. A. Walsh, J.-P. Bargetzi, and H. Neurath. Chemical relationships among various forms of bovine pancreatic carboxypeptidase A. Biochemistry 2:1475-1479, 1963.

885. Kumar, S., and K. S. J. Rao. Plasma and erythrocyte zinc levels in protein-calorie malnutrition. Nutr. Metabol. 15:364-371, 1973.

886. Kurus, E. Über den histochemischen Nachweis von Zink als Spurenelement in Auge des Menschen. Klin. Monatsbl. Augenheilkd. 134:338-350, 1959.

886a. Kurz, D. L., E. J. Eyring, and J. E. Roach. Serum zinc in the newborn. Biol. Neonate 23:180-183, 1973.

887. Kuykendall, W. E., Jr., L. E. Fite, and R. E. Wainerdi. Instrumental neutron activation analysis of air filter samples. J. Radioanal. Chem. 19:351-358, 1974.

888. Labbe, P., C. Volland, and P. Chaix. Étude de l'activité ferrochélatase des mitochondries de levure. Biochim. Biophys. Acta 159:527-539, 1968.

888a. LaBella, F. S., R. Dular, P. Lemon, S. Vivian, and G. Queen. Prolactin secretion is specifically inhibited by nickel. Nature 245:330-332, 1973.

888b. LaBella, F., R. Dular, S. Vivian, and G. Queen. Pituitary hormone releasing or inhibiting activity of metal ions present in hypothalamic extracts. Biochem. Biophys. Res. Commun. 52:785-791, 1973.

889. Lacko, A. G., and H. Neurath. Studies on procarboxypeptidase A and carboxypeptidase A of the spiny Pacific dogfish (Squalus acanthias). Biochemistry 9:4680-4690, 1970.

890. Lagerwerff, J. V., D. L. Brower, and G. T. Biersdorf. Accumulation of cadmium, copper, lead, and zinc in soil and vegetation in the proximity of a smelter, pp. 71-78. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - VI. Proceedings of 6th Annual Conference on Trace Substances in Environmental Health held in Columbia, Missouri, June 13-15, 1972. Columbia: University of Missouri, 1973.

891. Lagerwerff, J. V., and A. W. Specht. Contamination of roadside soil and vegetation with cadmium, nickel, lead and zinc. Environ. Sci. Technol. 4:583-586, 1970.

892. Lahav, N., and M. Hochberg. Kinetics of fixation of iron and zinc applied as FeEDTA, FeDDHA and ZnEDTA in the soil. Soil Sci. Soc. Amer. Proc. 39:55-58, 1975.

893. Lancaster, R. J., M. R. Coup, and J. W. Hughes. Toxicity of arsenic present in lakeweed. N. Z. Vet. J. 19:141-145, 1971.

894. Lander, D. W., R. L. Steiner, D. H. Anderson, and R. L. Dehm. Spectrographic determination of elements in airborne dirt. Appl. Spectrosc. 25:270-275, 1971.

895. Landry, A. S. The simultaneous determination of lead and zinc in atmospheric samples. A polarographic method utilizing a double internal standard. J. Ind. Hyg. Toxicol. 29:168-174, 1947.

896. Lantzsch, H.-J., and K. H. Menke. Untersuchungen zur Charakterisierung des Zn-Versorgungsstatus mit Hilfe von Chelatbildnern. Z. Tierphysiol. 32:129-143, 1973.

897. Lantzsch, H.-J., and K. H. Menke. Zur Bestimmung der intestinalen Sekretion und Absorption von Zink bei der wachsenden Ratte. Z. Tierphysiol. 34: 43-49, 1974.

898. Łapińska, K., and H. Łapiński. A case of parakeratosis in piglets. Med. Weter. 28:694, 1972. (in Polish)

899. Larsen, I. L., W. C. Renfro, and N. Cutshall. Zinc-65 specific activity in Mytilus californianus tissues, pp. 747-751. In D. J. Nelson, Ed. Radionuclides in Ecosystems. Vol. 2. Proceedings of the Third National Symposium on Radioecology, May 10-12, 1971, Oak Ridge, Tennessee. CONF-710501. Oak Ridge: U. S. Atomic Energy Commission, (not dated).

899a. Larson, D. L., M. Dobrkovsky, S. Abston, and S. R. Lewis. Zinc concentrations in plasma, red blood cells, wound exudate and tissues of burned children, pp. 627-632. In P. Matter, T. L. Barclay, and Z. Konickova, Eds. Research in Burns. Transactions of the Third International Congress, held in Prague Sept. 20-25, 1970. Bern: Hans Huber Publishers, 1971.

899b. Larson, D. L., R. Maxwell, S. Abston, and M. Dobrkovsky. Zinc deficiency in burned children. Plast. Reconstr. Surg. 46:13-21, 1970.

899c. Larson, D. L. Oral zinc sulfate in the management of severely burned patients, pp. 229-236. In W. J. Pories, W. H. Strain, J. M. Hsu, and R. L. Woolsey, Eds. Clinical Applications of Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1974.

900. Laurence, G. Zinc poisoning. Brit. Med. J. 1:582, 1958. (letter)

901. Laurent, G., M. Charrel, F. Luccioni, M. F. Autran, and J. Darrien. Sur les anhydrases carboniques erythrocytaires humaines. 1. Isolement et purification. Bull. Soc. Chim. Biol. 47:1101-1124, 1965.

902. Lavy, U. I. The effect of oral supplementation of zinc sulphate on primary wound healing in rats. Brit. J. Surg. 59:194-196, 1972.

903. Lawkowicz, W. P., I. I. Kszeminska-Lawkowicz, S. Szmigolski, and I. Litwin. Zinc metabolism in the cells of hemopoietic system in proliferative diseases. Fed. Proc. 25 (Translation Suppl.):809-810, 1966.

904. Lease, J. G. Effect of histidine on tibia alkaline phosphatase of chicks fed zinc-deficient sesame meal diets. J. Nutr. 102:1323-1330, 1972.

905. Lease, J. G. Effect of variations in dietary protein and amino acids on the alkaline phosphatase of the zinc-deficient chick. J. Nutr. 105:385-392, 1975.

906. Lease, J. G., B. D. Barnett, E. J. Lease, and D. E. Turk. The biological unavailability to the chick of zinc in a sesame meal ration. J. Nutr. 72:66-70, 1960.

907. Lease, J. G., and W. P. Williams, Jr. Availability of zinc and comparison of in vitro and in vivo zinc uptake of certain oil seed meals. Poult. Sci. 46:233-241, 1967.

907a. Leathem, J. H. Nutritional effects on hormone production. J. Anim. Sci. 25(Suppl.):68-82, 1966.

908. Leatherland, T. M., and J. D. Burton. The occurrence of some trace metals in coastal organisms with particular reference to the Solent region. J. Marine Biol. Assoc. U. K. 54:457-468, 1974.

909. Leaver, A. G. An effect of vitamin D upon the uptake and release of zinc by bone. Arch. Oral Biol. 12:773-775, 1967.

910. Ledbetter, J. O. Air sampling equipment, filters, pp. 214-233. In Air Pollution in Two Parts. Part A -- Analysis. New York: Marcel Dekker, Inc., 1972.

911. Lee, A. O., R. M. Jacobs, M. R. S. Fox, B. E. Fry, Jr., and D. Huisingh. The biological availability of zinc-65 from soybeans in Japanese quail. Fed. Proc. 34:907, 1975. (abstract)

912. Lee, C. R., G. R. Craddock, and H. E. Hammar. Factors affecting plant growth in high-zinc medium: I. Influence of iron on growth of flax at various zinc levels. Agron. J. 61:562-565, 1969.

913. Lee, C. R., and N. R. Page. Soil factors influencing the growth of cotton following peach orchards. Agron. J. 59:237-240, 1967.

914. Lee, R. E., Jr., S. S. Goranson, R. E. Enrione, and G. B. Morgan. National air surveillance cascade impactor network. II. Size distribution measurements of trace metal components. Environ. Sci. Technol. 6: 1025-1030, 1972.

914a. Lee, R. E., Jr., and D. J. von Lehmden. Trace metal pollution in the environment. J. Air Pollut. Control Assoc. 23:853-857, 1973.

915. Lee, K.-Y., and E. D. Weinberg. Sporulation of Bacillus megaterium: Roles of metal ions. Microbios 3:215-224, 1971.

916. Leeper, G. W. Reactions of Heavy Metals with Soils with Special Regard to Their Applications in Sewage Wastes. Prepared for Department of the Army, Corps of Engineers, under Contract No. DACW73-73-C-0026, 1972. 70 pp.

917. Leh, F., and K. M. Chan. Instruments and methods for determining trace metals. Chemtech 1974:178-182.

918. Lehman, G. S., and L. G. Wilson. Trace element removal from sewage effluent by soil filtration. Water Resources Res. 7:90-99, 1971.

918a. Lei, K. Y., A. Abbasi, and A. S. Prasad. Function of pituitary-gonadal axis in zinc-deficient rats. Amer. J. Physiol. 230:1730-1732, 1976.

919. Lehmann, K. B. Studien über technisch und hygienish wichtige Gase und Dämpfe. XIV. Das Giess oder Zinkfieber. Arch. Hyg. 72:358-381, 1910.

920. Leibetseder, J., A. Kment, M. Skalicky, H. Niedermüller, and G. Hofecker. Über den Zinkstoffwechsel beim Schwein. 3. Zinkgehalt und Dynamik des Zinkstoffwechsels der Organe. Wien. Tierärztl. Monatsschr. 59: 153-160, 1972.

921. Leland, H. V., E. D. Copenhaver, and L. S. Corrill. Heavy metals and other trace elements. J. Water Pollut. Control Fed. 46:1452-1476, 1974.

922. Lema, O., and H. H. Sandstead. Zinc deficiency: Effect on epiphyseal growth. Clin. Res. 18:458, 1970. (abstract)

923. Delete--use 922.

924. Leroux, J., and M. Mahmud. Flexibility of x-ray emission spectrography as adapted to microanalysis of air pollutants. J. Air Pollut. Control Assoc. 20:402-404, 1970.

925. Leutwein, F., and H. J. Rösler. Geochemische Untersuchungen an paläozoischen und mesozoischen Kohlen Mittel- und Ostdeutschlands. Freiberger Forsch. C19:1-196, 1956.

925a. Levine, J. B., R. Aamodt, D. P. Tschudy, and R. Henkin. Histadine-mediated decreases in total body zinc and in urinary porphyrins and porphyria. J. Clin. Invest. 52:52a, 1973. (abstract)

926. Levitt, J., and G. W. Todd. Metal-protein complexes in the potato. Physiol. Plant. 5:419-429, 1952.

927. Leyden, R. F., and S. J. Toth. Behavior of zinc sulfate as foliar applications and as soil applications in some New Jersey soils. Soil Sci. 89:223-228, 1960.

928. Li, T. K. The functional role of zinc in metalloenzymes, pp. 48-68. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

929. Li, T.-K., and B. L. Vallee. The biochemical and nutritional role of trace elements, pp. 372-399. In R. S. Goodhart and M. E. Shils, Eds. Modern Nutrition in Health and Disease. Dietotherapy. (5th ed.) Philadelphia: Lea & Febiger, 1973.

929a. Lichti, E. L., C. H. Almond, J. H. Henzel, and M. S. DeWeese. Differences in maternal and fetal plasma zinc levels in sheep and goats. Amer. J. Obstet. Gynecol. 106:1242-1244, 1970.

930. Lichti, E. L., J. A. Schilling, and H. M. Shurley. Wound fluid and plasma zinc levels in rats during tissue repair. Amer. J. Surg. 123:253-256, 1972.

931. Lichti, E. L., M. Turner, J. H. Henzel, and M. S. DeWeese. Wound fluid zinc levels during tissue repair. Sequential determination by means of surgically implanted teflon cylinders. Amer. J. Surg. 121:665-667, 1971.

931a. Lieberman, I., and P. Ove. Deoxyribonucleic acid synthesis and its inhibition in mammalian cells cultured from the animal. J. Biol. Chem. 237:1634-1642, 1962.

931b. Lieberman, I., R. Abrams, N. Hunt, and P. Ove. Levels of enzyme activity and deoxyribonucleic acid synthesis in mammalian cells cultured from the animal. J. Biol. Chem. 238:3955-3962, 1963.

932. Lifschitz, M. D., and R. I. Henkin. Circadian variation in copper and zinc in man. J. Appl. Physiol. 31:88-92, 1971.

933. Lin, Y. H., and J. R. Lawson. Treatment of oily and metal-containing waste-water. Pollut. Eng. 5(11):45-48, 1973.

933a. Lindeman, R. D., and D. J. Baxter. Zinc binding in serum and urine of cirrhotic, nephrotic, and normal subjects. J. Clin. Invest. 53:47a, 1974. (abstract)

934. Lindeman, R. D., R. G. Bottomley, R. L. Cornelison, Jr., and L. A. Jacobs. Influence of acute tissue injury on zinc metabolism in man. J. Lab. Clin. Med. 79:452-460, 1972.

935. Lindeman, R. D., M. L. Clark, J. P. Colmore. Influence of age and sex on plasma and red-cell zinc concentrations. J. Gerontol. 26:358-363, 1971.

936. Lindeman, R. D., A. A. Yunice, D. J. Baxter, L. R. Miller, and J. Nordquist. Myocardial zinc metabolism in experimental myocardial infarction. J. Lab. Clin. Med. 81:194-204, 1973.

936a. Lindholmer, C., and R. Eliasson. The effects of albumin, magnesium, and zinc on human sperm survival in different fractions of split ejaculates. Fertil. Steril. 25:424-431, 1974.

936b. Lindholmer, C., and R. Eliasson. In vitro release and uptake of zinc and magnesium by human spermatozoa. Int. J. Fertil. 19:56-62, 1974.

936c. Lindholmer, C., and R. Eliasson. Zinc and magnesium in human spermatozoa from different fractions of split ejaculates. Int. J. Fertil. 19: 45-48, 1974.

937. Lindholmer, C., and H. Glauman. Zinc and magnesium in human male reproductive tract. Andrologie 4:231-237, 1972.

938. Lindsay, W. Zinc in soils and plant nutrition. Adv. Agron. 24:147-186, 1972.

939. Lindsay, W. L. Inorganic phase equilibria of micronutrients in soils, pp. 41-57. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

940. Lindsay, W. L. Role of chelation in micronutrient availability, pp. 507-524. In E. W. Carson, Ed. The Plant Root and Its Environment. Proceedings of an Institute Sponsored by the Southern Regional Education Board, held at Virginia Polytechnic Institute and State University, July 5-16, 1971. Charlottesville: University Press of Virginia, 1974.

941. Lindsay, W. L., and W. A. Norvell. Equilibrium relationships of Zn^{2+}, Fe^{3+}, Ca^{2+}, and H^{+} with EDTA and DTPA in soils. Soil Sci. Soc. Amer. Proc. 33:62-68, 1969.

942. Lindskog, S., L. E. Henderson, K. K. Kannan, A. Liljas, P. O. Nyman, and B. Strandbert. Carbonic anhydrase, pp. 587-665. In P. D. Boyer, Ed. The Enzymes. Vol. 5. Hydrolysis. Sulfate Esters, Carboxyl Esters, Glycosides. Hydration. (3rd ed.) New York: Academic Press, 1971.

943. Linnenbom, V. J. Laboratory for the study of radioactive contamination of the sea (Fiascherino, Italy). Eur. Sci. Notes 27:296-298, 1973.

944. Lipsett, C. H. Industrial Wastes. Their Conservation and Utilization. New York: The Atlas Publishing Co., Inc., 1951. 317 pp.

945. Liptrap, D. O., E. R. Miller, D. E. Ullrey, D. L. Whitenack, B. L. Schoepke, and R. W. Luecke. Sex influence on the zinc requirement of developing swine. J. Anim. Sci. 30:736-741, 1970.

946. Little, P., and M. H. Martin. A survey of zinc, lead, and cadmium in soil and natural vegetation around a smelting complex. Environ. Pollut. 3:241-254, 1972.

947. Livingston, R. B., and P. A. Bentley. Role of Aquatic Vascular Plants in the Eutrophication of Selected Lakes in Western Massachusetts. Water Resources Research Center Publication No. 11. Amherst: University of Massachusetts, 1970. 80 pp.

948. Lloyd, R. Effect of dissolved oxygen concentrations on the toxicity of several poisons to rainbow trout (Salmo gairdnerii Richardson). J. Exp. Biol. 38:447-455, 1961.

948a. Lo, J. S., and J. A. Kellen. Spontaneous partial reactivation of chelated placental alkaline phosphatase in the absence of zinc. Enzyme 12:606-617, 1971.

948b. Lo, M.-C. Chemical application of diphenylthiocarbazone (dithizone) in carcinoma of the prostate. Can. Med. Assoc. J. 82:1203-1216, 1960.

949. Lohmander, S., U. Friberg, and B. Bergman. In vitro studies on the effect of in vivo zinc deficiency on the formation of glycosaminoglycans in rat costal cartilage. Acta Odontol. Scand. 30:89-96, 1972.

950. Lokken, P. M., E. S. Halas, and H. H. Sandstead. Influence of zinc deficiency on behavior. Proc. Soc. Exp. Biol. Med. 144:680-682, 1973.

950a. Lombeck, I., H. G. Schnippering, F. Ritzl, L. E. Feinendegen, and H. J. Bremer. Absorption of zinc in acrodermatitis enteropathica. Lancet 1:855, 1975. (letter)

950b. Lombeck, I., H. G. Schnippering, K. Kasperek, F. Ritzl, H. Kästner, L. E. Feinendegen, and H. J. Bremer. Akrodermatitis enteropathica--eine Zinkstoffwechselstörung mit Zinkmalabsorption. Z. Kinderheilkd. 120: 181-189, 1975.

951. Lopez, H., P. Isbell, J. Anderson, and J. M. Navia. The metabolism of zinc during bone fracture healing. J. Dent. Res. (Suppl.) 52:82, 1973. (abstract)

952. Lopez, P. L., and E. R. Graham. Labile pool and plant uptake of micronutrients: II. Uptake of Mn, Fe, and Zn by Ladino clover (Trifolium repens) and its relation to soil labile pools. Soil Sci. 115:380-389, 1973.

952a. Delete--use 526a.

953. Los, L. I., L. K. Pyatnitskaya, and A. S. Samsonova. Further observations over the content of certain trace elements in foodstuffs of plant origin. Vopr. Pitan. 25(2):84-85, 1966. (in Russian)

954. Lucas, H. F., Jr., D. N. Edgington, and P. J. Colby. Concentrations of trace elements in Great Lakes fishes. J. Fish. Res. Board Can. 27: 677-684, 1970.

955. Lucas, R. E. Micronutrients for Vegetables and Field Crops. Extension Bulletin E-486. Farm Science Series. Cooperative Extension Service. Michigan State University, 1964. 12 pp.

956. Lucas, R. E., and B. D. Knezek. Climatic and soil conditions promoting micronutrient deficiencies in plants, pp. 265-288. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

957. Luecke, R. W. The significance of zinc in nutrition. Borden's Rev. Nutr. Res. 26:45-53, 1965.

958. Luke, C. L., T. Y. Kometani, J. E. Kessler, T. C. Loomis, J. L. Bove, and B. Nathanson. X-ray spectrometric analysis of air pollution dust. Environ. Sci. Technol. 6:1105-1109, 1972.

958a. Lutwak-Mann, C., and J. E. A. McIntosh. Zinc and carbonic anhydrase in rabbit uterus. Nature 221:1111-1114, 1969.

958b. Lum, K. L. L., and R. I. Henkin. Sugar binding to purified fractions from bovine taste buds and epithelial tissue. Biochim. Biophys. Acta 421: 380-394, 1976.

958c. Lum, C. K. L., and R. I. Henkin. Characterization of fractions from taste bud and non-taste bud-enriched filtrates from and around bovine circumvallate papillae. Biochim. Biophys. Acta 421:362-379, 1976.

958d. Lum, K. L. L., N. F. Whittaker, and R. I. Henkin. Preparation and isolation of a taste bud-derived fraction from bovine circumvallate papillae. Biochim. Biophys. Acta 421:353-361, 1976.

959. Horvick, E. W. Zinc and zinc alloys, pp. 1157-1172. In T. Lyman, Ed. Metals Handbook. Vol. 1. Properties and Selection of Metals. (8th ed.) Metals Park, Novelty, Ohio: American Society for Metals, Inc., 1961.

960. Maas, E. V., G. Ogata, and M. J. Garber. Influence of salinity on Fe, Mn, and Zn uptake by plants. Agron. J. 64:793-795, 1972.

961. Macapinlac, M. P., G. H. Barney, W. M. Pearson, and W. J. Darby. Production of zinc deficiency in the squirrel monkey (Saimiri sciureus). J. Nutr. 93:499-510, 1967.

962. Macapinlac, M. P., W. N. Pearson, G. H. Barney, and W. J. Darby. Protein and nucleic acid metabolism in the testes of zinc-deficient rats. J. Nutr. 95:569-577, 1968.

963. MacGregor, J. M., A. Sajjapongse, and O. M. Gunderson. Availability of fertilizer zinc to corn in a calcareous mineral soil. Soil Sci. Soc. Amer. Proc. 38:611-616, 1974.

964. Mackenzie, A. R., T. Hall, and W. F. Whitmore, Jr. Zinc content of expressed human prostatic fluid. Nature 193:72-73, 1962.

965. Mackenzie, A. R., T. Hall, M.-C. Lo., and W. F. Whitmore, Jr. Influence of castration and sex hormones on size, histology and zinc content of canine prostate. J. Urol. 89:864-874, 1963.

966. MacMahon, R. A., L. J. Cussen, F. T. McDermott, and B. E. James. Thymidine incorporation in esophageal wounds of zinc-deficient rats. Surgery 75: 660-663, 1974.

967. Macy, I. G., H. J. Kelly, and R. E. Sloan. The Composition of Milks. A Compilation of the Comparative Composition and Properties of Human, Cow, and Goat Milk, Colostrum, and Transitional Milk. NAS Publ. 254. Washington, D. C.: National Academy of Sciences, 1953. 70 pp.

968. Magee, A. C., M. Y. Jackson, and W. Wade. Interrelationships between copper, iron and zinc in young rats fed zinc deficient diets. Fed. Proc. 34:906, 1975. (abstract)

969. Magee, A. C., and G. Matrone. Studies on growth, copper metabolism and iron metabolism of rats fed high levels of zinc. J. Nutr. 72:233-242, 1960.

970. Mahanand, D., and J. C. Houck. Fluorometric determination of zinc in biological fluids. Clin. Chem. 14:6-11, 1968.

971. Mahlen, D. J., J. R. Walsh, and G. D. Haynie. Magnesium, zinc and copper in dialyses patients. Amer. J. Clin. Path. 56:17-23, 1971.

971a. Mallette, L. E., J. P. Bilezekian, D. A. Heath, and G. D. Aurbach. Primary hyperparathyroidism: Clinical and biochemical features. Medicine 53:127-146, 1974.

971b. Mallette, L. E., and R. I. Henkin. Altered copper and zinc metabolism in primary hyperparathyroidism. Amer. J. Med. Sci. 272:167-174, 1976.

971c. Mallette, L. E., and R. I. Henkin. Increased urinary Cu and Zn excretion in primary hyperparathyroidism. Clin. Res. 24:364a, 1976. (abstract)

972. Makdani, D., O. Mickelsen, D. Ullrey, and P. K. Ku. Effect of phytic acid on rat growth and availability of zinc, copper, iron and calcium. Fed. Proc. 34:926, 1975. (abstract)

972a. Markkaren, T. Metabolic disturbance after gastro-oesophageal resection. Int. J. Vit. Nutr. Res. 43:549-554, 1973.

972b. Makotchenko, V. M. The functional condition of suprarenal cortex in chronic poisoning with heavy metals (lead, mercury). Tr. Ukr. Nauch. Issled Inst. Eksp. Endokrinol. 20:162-170, 1965. (in Russian)

973. Mann, S. O., B. F. Fell, and A. C. Dalgarno. Observations on the bacterial flora and pathology of the tongue of sheep deficient in zinc. Res. Vet. Sci. 17:91-101, 1974.

973a. Mann, T. The Biochemistry of Semen and of the Male Reproductive Tract. New York: John Wiley & Sons Inc., 1964. pp. 90-91.

974. Mann, T., and D. Keinlin. Haemocuprein and hepatocuprein, copper-protein compounds of blood and liver in mammals. Proc. Roy. Soc. B 126: 303-315, 1939.

975. Mansouri, K., J. A. Halsted, and E. A. Gombos. Zinc, copper, magnesium and calcium in dialyzed and non-dialyzed uremic patients. Arch. Intern. Med. 125:88-93, 1970.

976. Manzler, A. D., and A. W. Schreiner. Copper-induced acute hemolytic uremia. A new complication of hemodialysis. Ann. Intern. Med. 73:409-412, 1970.

977. Marinho, M. L., and K. Igue. Factors affecting zinc absorption of corn from volcanic ash soils. Agron. J. 64:3-8, 1972.

978. Marshall, B. S., I. Telford, and R. Wood. A field method for the determination of zinc oxide fume in air. Analyst 96:569-578, 1971.

979. Martens, D. C., M. T. Carter, and G. D. Jones. Response of soybeans following six annual applications of various levels of boron, copper, and zinc. Agron. J. 66:82-84, 1974.

980. Martens, D. C., G. W. Hawkins, and G. D. McCart. Field response of corn to $ZnSO_4$ and Zn-EDTA placed with the seed. Agron. J. 65:135-136, 1973.

981. Martinez, A., and D. C. Church. Effect of various mineral elements on *in vitro* rumen cellulose digestion. J. Anim. Sci. 31:982-990, 1970.

982. Mashar, U. Über teratoide Geschwülste in der Leibeshöle beim Haushuhn. Virchows Arch. Path. Anat. Physiol. Klin. Med. 285:155-168, 1932.

982a. Masironi, R. Trace elements and cardiovascular disease. Bull. World Health Org. 40:305-312, 1969.

983. Mason, K. E., and J. O. Young. Effectiveness of selenium and zinc in protecting against cadmium-induced injury of the rat testis, pp. 383-394. In O. H. Muth, Ed. Symposium: Selenium in Biomedicine. First International Symposium, Oregon State University, 1966. Westport, Conn.: The AVI Publishing Company, Inc., 1967.

984. Massey, H. F., and F. A. Loeffel. Factors in interstrain variation in zinc content of maize (*Zea mays* L.) kernels. Agron. J. 59:214-217, 1967.

985. Mathies, J. C. Preparation and properties of highly purified alkaline phosphatase from swine kidneys. J. Biol. Chem. 233:1121-1227, 1958.

985a. Mathies, J. C., and P. K. Lund. X-ray spectrographic microanalysis of human tissues for iron and zinc. Amer. J. Clin. Path. 59:417-422, 1973.

986. Matoušek, J. P., and K. G. Brodie. Direct determination of lead airborne particulates by nonflame atomic absorption. Anal. Chem. 45:1606-1609, 1973.

987. Matrone, G. Chemical parameters in trace-element metabolism, pp. 91-103. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium on Trace Element Metabolism in Animals, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

988. Matson, W. R. Trace Metals, Equilibrium and Kinetics of Trace Metal Complexes in Natural Media. Ph.D. Thesis. Cambridge: Massachusetts Institute of Technology, 1968. 272 pp.

989. Matson, W. R., R. M. Griffin, and G. B. Schreiber. Rapid sub-nanogram simultaneous analysis of Zn, Cd, Pb, Cu, Bi, and Tl, pp. 396-406. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - IV. Proceedings of University of Missouri's 4th Annual Conference on Trace Substances in Environmental Health held in Columbia, Missouri, June 23-25, 1970. Columbia: University of Missouri, 1971.

989a. Matter, B. J., J. Pederson, G. Psimenos, and R. D. Lindeman. Lethal Cu intoxication in hemodialysis. Trans. Amer. Soc. Artif. Intern. Organs 15:309-315, 1969.

990. Matthews, B. W., J. N. Jansonius, P. M. Colman, B. P. Schoenborn, and D. Dupourque. Three-dimensional structure of thermolysin. Nature New Biol. 238:37-41, 1972.

991. Matzku, S., and E. Broda. Die Zinkaufnahme in das Innere von Chlorella. Planta 92:29-40, 1970.

992. Maust, L. E., W. G. Pond, and M. L. Scott. Energy value of a cassava-rice bran diet with and without supplemental zinc for growing pigs. J. Anim. Sci. 35:953-957, 1972.

993. Mawson, C. A., and M. I. Fischer. The occurrence of zinc in the human prostate gland. Can. J. Med. Sci. 30:336-339, 1952.

994. Mawson, C. A., and M. I. Fischer. Zinc content of the genital organs of the rat. Nature 167:859, 1951.

995. Maxwell, L. C. The quantitative and qualitative ovarian response to distributed dosage with gonadotropic extracts. Amer. J. Physiol. 110:458-463, 1934.

996. Mayer, J. Zinc deficiency: A cause of growth retardation? Postgrad. Med. 35:206-209, 1964.

997. Maynard, J. R., and J. E. Coleman. Elasmobranch carbonic anhydrase. Purification and properties of the enzyme from two species of shark. J. Biol. Chem. 246:4455-4464, 1971.

997a. Maxwell, L. C. The quantitative and qualitative ovarian response to distributed dosage with gonadotropic extracts. Amer. J. Physiol. 110:458-463, 1934.

998. Mazanowska, A. M., A. Neuberger, and G. H. Tait. Effect of lipids and organic solvents on the enzymic formation of zinc protoporphyrin and haem. Biochem. J. 98:117-127, 1966.

998a. MacMahon, R. A., M. L. M. Parker, and M. C. McKinnon. Zinc treatment in malabsorption. Med. J. Austral. 2:210-212, 1968.

999. McBean, L. D., M. Mahloudji, J. G. Reinhold, and J. A. Halsted. Correlation of zinc concentrations in human plasma and hair. Amer. J. Clin. Nutr. 24:506-509, 1971.

999a. McBean, L. D., J. C. Smith, Jr., B. H. Berne, and J. A. Halsted. Serum zinc and alpha$_2$-macroglobulin concentration in myocardial infarction, decubitus ulcer, multiple myeloma, prostatic carcinoma, Down's syndrome and nephrotic syndrome. Clin. Chim. Acta 50:43-41, 1974.

1000. McBean, L. D., J. C. Smith, Jr., and J. A. Halsted. Effect of oral contraceptive hormones on zinc metabolism in the rat. Proc. Soc. Exp. Biol. Med. 137:543-547, 1971.

1001. McBean, L. D., J. C. Smith, Jr., and J. A. Halsted. Zinc deficiency in guinea pigs. Proc. Soc. Exp. Biol. Med. 140:1207-1209, 1972.

1001a. McCabe, L. J. Problem of trace metals in water supply--an overview, pp. 1-9. In Proceedings. Sixteenth Water Quality Conference. Trace Metals in Water Supplies: Occurrence, Significance, and Control. University of Illinois, Champaign, Feb. 12-13, 1974.

1001b. McCabe, L. J., J. M. Symons, R. D. Lee, and G. G. Robeck. Survey of community water supply systems. J. Amer. Water Works Assoc. 62: 670-687, 1970.

1002. McClain, P. E., E. R. Wiley, G. R. Beecher, W. L. Anthony, and J. M. Hsu. Influence of zinc deficiency on synthesis and crosslinking of rat skin collagen. Biochim. Biophys. Acta 304:457-465, 1973.

1003. McConnell, K. P., J. M. Hsu, J. L. Herrman, and W. L. Anthony. Parallelism between sulfur and selenium amino acids in protein synthesis in the skin of zinc deficient rats. Proc. Soc. Exp. Biol. Med. 145:970-974, 1974.

1004. McConnell, S. D., and R. I. Henkin. Altered preference for sodium chloride, anorexia, and changes in plasma and urinary zinc in rats fed a zinc-deficient diet. J. Nutr. 104:1108-1114, 1974.

1005. McCord, C. P. Metal fume fever as an immunological disease. Ind. Med. Surg. 29:101-107, 1960.

1006. McCord, C. P., A. Friedlander, W. E. Brown, and D. K. Minster. An occupational disease among zinc workers. Arch. Intern. Med. 37:641-659, 1926.

1007. McCord, J. M., and I. Fridovich. Superoxide dismutase. An enzymic function for erythrocuprein (hemocuprein). J. Biol. Chem. 244:6049-6055, 1969.

1008. McCuaig, L. W., and I. Motzok. Interactions of Ca, P, Zn, and alkaline phosphatase in the chick. II. Effect of dietary Ca level. Can. J. Physiol. Pharmacol. 52:90-95, 1974.

1008a. McDonald, D. F. Effect of diphenyldithiocarbazone on prostates of animals and in human prostatic cancer. J. Urol. 83:458-462, 1960.

1009. McDonald, A. L., and N. W. Heimstra. Agonistic behavior in several species of fish. Psychol. Rep. 16:845-850, 1965.

1009a. McFarren, E. F., J. E. Campbell, and J. B. Engle. The occurrence of copper and zinc in shellfish, pp. 229-234. In E. T. Jensen, Ed. Proceedings. Shellfish Sanitation Workshop, November 28-30, 1961. Washington, D. C.: U. S. Department of Health, Education, and Welfare, Public Health Service, 1961.

1010. McFarren, E. F., J. H. Parker, and R. J. Lishka. Water Metals No. 4, Study No. 30. Report of a Study Conducted by Analytical Reference Service. Public Health Service Publ. No. 999-UIH-8. Cincinnati: U. S. Department of Health, Education, and Welfare. Public Health Service. Bureau of Disease Prevention and Control. 1968. 128 pp.

1011. McIntosh, J. E. A., and C. Lutwak-Mann. Zinc transport in rabbit tissues. Some hormonal aspects of the turnover of zinc in female reproductive organs, liver and body fluids. Biochem. J. 126:869-876, 1972.

1011a. McIntosh, J. E. A., and C. Lutwak-Mann. Zinc in luteal and interstitial tissue of the rabbit ovary in early pregnancy. Nature New Biol. 236:53-54, 1972.

1011b. McKee, J. E., and H. W. Wolf, Eds. Water Quality Criteria. (2nd ed.) Publication 3-A. Sacramento: California State Water Resources Control Board, 1963. 548 pp.

1012. McKenzie, J. M., G. J. Fosmire, and H. H. Sandstead. Zinc deficiency during the latter thrid of pregnancy: Effects on fetal rat brain, liver, and placenta. J. Nutr. 105:1466-1475, 1975.

1013. McKenzie, J. M., and D. L. Kay. Urinary excretion of cadmium, zinc and copper in normotensive and hypertensive women. N. Z. Med. J. 78: 68-70, 1973.

1014. McKenzie, J. M., and R. M. Pettit. Alteration of the zinc and copper concentration of hair. Nutrition Department, University of Otago School of Home Science, Dunedin, New Zealand, 1973.

1015. McKenzie, J. M., and R. M. Pettit. Zinc and copper concentrations in hair of some New Zealand men and women. Nutrition Department, University of Otago, School of Home Science, Dunedin, New Zealand, 1973.

1015a. McLaughlan, J. M., J. D. Jones, B. G. Shah, and J. L. Beare-Rogers. Reproduction in rats fed protein concentrate from mustard or rapeseed. Nutr. Rep. Int. 11:327-335, 1975.

1016. McMahon, A. D., C. H. Cotterill, J. T. Dunham, and W. L. Rice. The U. S. Zinc Industry: A Historical Perspective. U. S. Bureau of Mines. Information Circular 8629. Washington D. C.: U. S. Department of the Interior, 1974.

1016a. McMahon, A. D., J. M. Hague, and H. R. Babitzke. Zinc, pp. 1299-1333. In Minerals Yearbook. 1972. Vol. 1. Metals, Minerals and Fuels. Washington, D. C.: U. S. Department of the Interior, 1974.

1017. McNabb, C. D., Jr., and D. P. Tierney. Growth and Mineral Accumulation of Submersed Vascular Hydrophytes in Pleioeutrophic Environs. Technical Report No. 26. Institute of Water Resources. East Lansing: Michigan State University, 1972. 33 pp.

1018. McQuitty, J. T., Jr., W. D. DeWys, L. Monaco, W. H. Strain, C. G. Rob, J. Apgar, and W. J. Pories. Inhibition of tumor growth by dietary zinc deficiency. Cancer Res. 30:1387-1390, 1970.

1019. Mealey, M. Hydrometallurgy plays big role in Japan's new zinc smelter. Eng. Min. J. 174(1):82-84, 1973.

1020. Mechanisms of copper absorption in the chick. Nutr. Rev. 27:208-211, 1969.

1021. Medel, M., C. Espinoza, J. Zipper, and R. Prager. Preliminary observations on the effects of copper, zinc and polyethylene I.U.F.Bs. on the uterine motility of the rabbit. Contraception 5:203-213, 1972.

1022. Medel, M., C. Espinoza, J. Zipper, and R. Prager. Reversibility of the contraceptive action of copper and zinc in the rat and rabbit. Contraception 6:241-247, 1972.

1023. Meiggs, T. O. Report on water: General referee reports, Subcommittee D. J. Assoc. Off. Anal. Chem. 57:295, 1974.

1024. Mellinger, P. J. The Comparative Metabolism of Cadmium, Mercury and Zinc as Environmental Contaminants in the Freshwater Mussel, Margaritifera margaritifera. Ph.D. Thesis. Corvallis: Oregon State University, 1972. 129 pp.

1024a. Mellinkoff, S. M., M. Frankland, and M. Greipel. Effect of amino acid and glucose ingestion on arteriovenous blood sugar and appetite. J. Appl. Physiol. 9:85-87, 1956.

1024b. Mellinkoff, S. M., M. Frankland, D. Boyle, and M. Greipel. Relationship between serum amino acid concentration and fluctuations in appetite. J. Appl. Physiol. 8:535-538, 1956.

1025. Mellor, J. W. The history of zinc and cadmium, pp. 298-405. In A Comprehensive Treatise on Inorganic and Theoretical Chemistry. Vol. IV. Ra and Ac Families, Be, Mg, Zn, Cd, Hg. London: Longmans, Green and Co. Ltd., 1929.

1026. Melsted, S. W., H. L. Motto, and T. R. Peck. Critical plant nutrient composition values useful in interpreting plant analysis data. Agron. J. 61:17-20, 1969.

1027. Melton, J. R., B. G. Ellis, and E. C. Doll. Zinc, phosphorus, and lime interactions with yield and zinc uptake by Phaseolus vulgaris. Soil Sci. Soc. Amer. Proc. 34:91-93, 1970.

1028. Melton, J. R., S. K. Mahtab, and A. R. Swoboda. Diffusion of zinc in soils as a function of applied zinc, phosphorus, and soil pH. Soil Sci. Soc. Amer. Proc. 37:379-381, 1973.

1029. Menzel, R. G., and M. L. Jackson. Determination of copper and zinc in soil or plants. Anal. Chem. 23:1861-1863, 1951.

1030. Mercer, T. T., P. E. Morrow, and W. Stöber, Eds. Assessment of Airborne Particles. Fundamentals, Applications, and Implications to Inhalation Toxicity. Springfield, Ill.: Charles C Thomas, 1972. 540 pp.

1031. Meret, S., and R. I. Henkin. Simultaneous direct estimation by atomic absorption spectrophotometry of copper and zinc in serum, urine and cerebrospinal fluid. Clin. Chem. 17:369-373, 1971.

1032. Merlini, M. The freshwater clam as a biological indicator of radiomanganese, pp. 977-982. In B. Åberg and F. P. Hungate, Eds. Radioecological Concentration Processes. Proceedings of an International Symposium held in Stockholm 25-29 April, 1966. New York: Pergamon, 1967.

1033. Mertz, W. Human requirements: Basic and optimal. Ann. N. Y. Acad. Sci. 199:191-201, 1972.

1034. Mesrobian, A. Z., and G. Shklar. The effect of dietary zinc sulfate supplements on the healing of experimental extraction wounds. Oral Surg. Oral Med. Oral Path. 28:259-265, 1969.

1035. Methfessel, A. H., and H. Spencer. Zinc metabolism in the rat. I. Intestinal absorption of zinc. J. Appl. Physiol. 34:58-62, 1973.

1036. Methfessel, A. H., and H. Spencer. Zinc metabolism in the rat. II. Secretion of zinc into intestine. J. Appl. Physiol. 34:63-67, 1973.

1037. Meyer, J., and O. F. Alvares. Dry weight and size of cells in the buccal epithelium of zinc-deficient rats: A quantitative study. Arch. Oral Biol. 19:471-476, 1974.

1037a. Michaëlsson, G. Zinc therapy in acrodermatitis enteropathica. Acta Derm. Venereol. (Stockh.) 54:377-381, 1974.

1037b. Michaëlsson, G. Zinc--new and successful therapy for acrodermatitis enteropathica. Läkartidningen 71:1959-1961, 1974. (in Swedish, summary in English)

1038. Michalowsky, I. Das 10. experimentalle Zink-Teratoma. Virchows Arch. Path. Anat. Physiol. Klin. Med. 274:319-325, 1930.

1039. Michalowsky, I. Die experimentalle Erzeugung einer teratoiden Neubildung der Hoden beim Hahn. Zentralbl. Allg. Path. 38:585-587, 1926.

1040. Michalowsky, I. Eine experimentelle Erzeugung teratoider Geschwülste der Hoden beim Hahn. Virchows Arch. Path. Anat. Physiol. Klin. Med. 267:27-62, 1928.

1041. Micheli, A., and C. Buzzi. Radio-immunoelectrophoresis of red cell carbonic anhydrase labelled with ^{65}Zn. Biochim. Biophys. Acta 96:533-534, 1967.

1042. Michell, H. The Economics of Ancient Greece. (2nd ed.) New York: Barnes and Noble, Inc., 1957. pp. 118-119.

1043. Miesch, A. T., and C. Huffman, Jr. Abundance and distribution of lead, zinc, cadmium, and arsenic in soils, pp. 65-80. In Helena Valley, Montana, Area Environmental Pollution Study. Office of Air Programs Publication No. AP-91. Research Triangle Park, N. C.: U. S. Environmental Protection Agency, 1972.

1044. Mikac-Dević, D. Methodology of zinc determinations and the role of zinc in biochemical processes. Adv. Clin. Chem. 13:271-333, 1970.

1044a. Delete

1044b. Millar, M. J., M. I. Fischer, P. V. Elcoate, and C. A. Mawson. The effects of dietary zinc deficiency on the reproductive system of male rats. Can. J. Biochem. Physiol. 36:557-569, 1958.

1044c. Millar, M. J., P. V. Elcoate, M. I. Fischer, and C. A. Mawson. Effect of testosterone and gonadotrophin injections on the sex organ development of zinc-deficient male rats. Can. J. Biochem. Physiol. 38:1457-1466, 1960.

1045. Mildvan, A. S. Metals in enzyme catalysis, pp. 445-536. In P. D. Boyer, Ed. The Enzymes. Vol. 2. Kinetics and Mechanism. (3rd ed.) New York: Academic Press, 1970.

1045a. Miller, D. W., R. J. Vetter, R. L. Hullinger, and S. M. Shaw. Uptake and distribution of cadmium-115m in calcium deficient and zinc deficient golden hamsters. Bull. Environ. Contam. Toxicol. 13: 40-43, 1975.

1045b. Millar, M. J., M. I. Fischer, P. V. Elcoate, and C. A. Mawson. The effects of dietary zinc deficiency on the reproductive system of male rats. Can. J. Biochem. Physiol. 36:557-569, 1958.

1046. Miller, J. K., and R. G. Cragle. Gastrointestinal sites of absorption and endogenous secretion of zinc in dairy cattle. J. Dairy Sci. 48: 370-373, 1965.

1047. Miller, M. L., L. Murthy, C. R. Basom, and H. G. Petering. Alterations in hepatocytes after manipulation of the diet: Copper, zinc and cadmium interactions. Amer. J. Anat. 141:23-40, 1974.

1048. Miller, W. J. Dynamics of absorption rates, endogenous excretion, tissue turnover, and homeostatic control mechanisms of zinc, cadmium, manganese, and nickel in ruminants. Fed. Proc. 32:1915-1920. 1973.

1049. Miller, W. J. Zinc metabolism in farm animals, pp. 23-41. In Mineral Studies with Isotopes in Domestic Animals. Proceedings of a Panel on the Use of Nuclear Techniques in Studies of Mineral Metabolism and Disease in Domestic Animals, Vienna, 28 Sept.-2 Oct, 1970. Vienna: International Atomic Energy Agency, 1971.

1050. Miller, W. J. Zinc nutrition of cattle: A review. J. Dairy Sci. 53: 1123-1135, 1970.

1051. Miller, W. J., D. M. Blackmon, R. P. Gentry, and F. M. Pate. Effects of high but nontoxic levels of zinc in practical diets on ^{65}Zn and zinc metabolism in Holstein calves. J. Nutr. 100:893-902, 1970.

1052. Miller, W. J., D. M. Blackmon, R. P. Gentry, and F. M. Pate. Zinc distribution in various tissues of zinc-deficient and normal bull calves with time after single intravenous or oral dosing. J. Anim. Sci. 31: 149-156, 1970.

1053. Miller, W. J., D. M. Blackmon, R. P. Gentry, W. J. Pitts, and G. W. Powell. Absorption, excretion and retention of orally administered zinc-65 in various tissues of zinc-deficient and normal goats and calves. J. Nutr. 92:71-78, 1967.

1054. Miller, W. J., D. M. Blackmon, J. M. Hiers, Jr., P. R. Fowler, C. M. Clifton, and R. P. Gentry. Effects of adding two forms of supplemental zinc to a practical diet on skin regeneration in Holstein heifers and evaluation of a procedure for determining rate of wound healing. J. Dairy Sci. 50:715-521, 1967.

1054a. Miller, J. K., and W. J. Miller. Experimental zinc deficiency and recovery of calves. J. Nutr. 76:467-474, 1962.

1055. Miller, W. J., and J. K. Miller. Photomicrographs of skin from zinc-deficient calves. J. Dairy Sci. 46:1285-1287, 1963.

1056. Miller, W. J., and J. K. Miller. Zinc content of certain feeds, associated materials and water. J. Dairy Sci. 46:581-583, 1963.

1057. Miller, W. J., and J. K. Miller. Zinc content of some feed ingredients and material. J. Dairy Sci. 46:366-367, 1963. (abstract)

1058. Miller, W. J., J. D. Morton, W. J. Pitts, and C. M. Clifton. Effect of zinc deficiency and restricted feeding on wound healing in the bovine. Proc. Soc. Exp. Biol. Med. 118:427-430, 1965.

1059. Miller, W. J., G. W. Powell, D. M. Blackmon, and R. P. Gentry. Zinc and dry matter content of tissues and feces of zinc-deficient and normal ruminants fed ethylenediaminetetraacetate and cadmium. J. Dairy Sci. 51:82-89, 1968.

1060. Miller, W. J., E. S. Wells, R. P. Gentry, and M. W. Neathery. Endogenous zinc excretion and ^{65}Zn metabolism in Holstein calves fed intermediate to high but nontoxic zinc levels in practical diets. J. Nutr. 101: 1673-1682, 1971.

1061. Millikan, C. R. Effect of molybdenum on the severity of toxicity symptoms in flax induced by an excess of either manganese, zinc, copper, nickel, or cobalt in the nutrient solution. J. Austral. Inst. Agric. Sci. 13:180-186, 1947.

1062. Millikan, C. R., and B. C. Hanger. Effects of chelation and of various cations on the mobility of foliar-applied ^{65}Zn in subterranean clover. Austral. J. Biol. Sci. 18:953-957, 1965.

1062a. Mills, C. F. The detection of trace element deficiency and excess in man and farm animals. Proc. Nutr. Soc. 33:267-274, 1974.

1063. Mills, C. F., and A. C. Dalgarno. Copper and zinc status of ewes and lambs receiving increased dietary concentrations of cadmium. Nature 239:171-173, 1972.

1064. Mills, C. F., A. C. Dalgarno, R. B. Williams, and J. Quarterman. Zinc deficiency and the zinc requirements of calves and lambs. Brit. J. Nutr. 21:751-768, 1967.

1065. Mills, C. F., J. Quarterman, J. K. Chesters, R. B. Williams, and A. C. Dalgarno. Metabolic role of zinc. Amer. J. Clin. Nutr. 22:1240-1249, 1969.

1066. Mills, C. F., J. Quarterman, R. B. Williams, and A. C. Dalgarno. The effects of zinc deficiency on pancreatic carboxypeptidase activity and protein digestion and absorption in the rat. Biochem. J. 102: 712-718, 1967.

1067. Mink, L. L. R. E. Williams, and A. T. Wallace. Analysis of an aquatic environment receiving domestic and industrial effluent, pp. 69-84. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - IV. Proceedings of University of Missouri's 4th Annual Conference on Trace Substances in Environmental Health held in Columbia, Missouri, June 23-24, 1970. Columbia: University of Missouri, 1971.

1068. Mink, L. L., R. E. Williams, and A. T. Wallace. Effect of industrial and domestic effluents on the water quality of the Coeur d'Alene River basin, 1969-1970. Idaho Bureau of Mines and Geology Pamphlet 149:1-30, 1971.

1069. Mishima, J., and E. P. Odum. Excretion rate of Zn^{65} by Littorina irrorata in relation to temperature and body size. Limnol. Oceanogr. 8:39-44, 1963.

1070. Misra, H. P., and I. Fridovich. The purification and properties of superoxide dismutase from Neurospora crassa. J. Biol. Chem. 247:3410-3414, 1972.

1071. Mitsugi, H., N. Takata, M. Motoyama, M. Akamatsu, and G. Hashizume. Determination of zinc and lead in suspended particulates by fluorescent x-ray spectrometry. Bunseki Kagaku (Jap. Anal.) 19:1383-1388, 1970. (in Japanese)

1072. Miura, Y., and S. Tsunashima. Incorporation of zinc-65 into goldfish. Kagaku (Science) 26:314-315, 1956. (in Japanese)

1073. Mizohata, A., and T. Mamuro. Elemental analysis of airborne dust by energy dispersive fluorescent x-ray spectrometry. Ann. Rep. Radiat. Cent. Osaka Prefect. 13:16-22, 1972.

1074. Mizohata, A., and T. Mamuro. Elemental analysis of airborne dust by energy dispersive fluorescent x-ray spectrometry (II). Ann. Rep. Radiat. Cent. Osaka Prefect. 14:19-22, 1973.

1075. Mizuno, T. Observations on the behavior of zinc during the course of spermatogenesis in the grasshopper, Atractomorpha bedeli. J. Faculty Sci. Univ. Tokyo (Tokyo Daigaku Rigakubu) 8(Sect. 4):211-217, 1958.

1075a. Molokhia, M. M., and B. Portnoy. Neutron activation analysis of trace elements in skin. III. Zinc in normal skin. Brit. J. Derm. 81: 759-762, 1969.

1075b. Mahler, D. J., J. R. Walsh, and G. D. Haynie. Magnesium, zinc and copper in dialysis patients. Amer. J. Clin. Path. 56:17-23, 1971.

1076. Moncilovic, B., B. Belonje, and B. G. Shah. The choice of young rat tissue for a zinc bioassay. Fed. Proc. 34:906, 1975. (abstract)

1077. Monier-Williams, G. W. Trace Elements in Food. (2nd ed.) New York: John Wiley & Sons, 1949. 511 pp.

1078. Moore, D. P. Mechanisms of micronutrient uptake by plants, pp. 171-198. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

1079. Morgan, J. M., H. B. Burch, and J. B. Watkins. Tissue cadmium and zinc content in emphysema and brochogenic carcinoma. J. Chron. Dis. 24: 107-110, 1971.

1080. Morgan, R. S., N. H. Morgan, and R. A. Guinavan. Zinc in Sephadex. Anal. Biochem. 45:668-669, 1972.

1081. Mortensen, J. L. Complexing of metals by soil organic matter. Soil Sci. Soc. Amer. Proc. 27:179-186, 1963.

1082. Morton, J. J. P., and M. H. Malone. Evaluation of vulnerary activity by an open wound procedure in rats. Arch. Int. Pharmacodyn. Ther. 196:117-126, 1972.

1083. Moss, D. W. The influence of metal ions on the orthophosphatase and inorganic pyrophosphatase activities of human alkaline phosphatase. Biochem. J. 112:699-701, 1969.

1084. Motsara, M. R. On the effects of phosphorus on zinc uptake by barley. Plant Soil 38:381-392, 1973.

1085. Moynahan, E. J. Acrodermatitis enteropathica: A lethal inherited human zinc-deficiency disorder. Lancet 2:399-400, 1974.

1085a. Moynahan, E. J. Zinc deficiency and disturbances of mood and visual behaviour. Lancet 1:91, 1976. (letter)

1085b. Moynahan, E. J. Acrodermatitis enteropathica with secondary lactose intolerance, and tertiary deficiency state, probably due to chelation of essential nutrients by di-iodhydroxyquinolone. Proc. Roy. Soc. Med. 59:445-447, 1966.

1086. Moynahan, E. J., and P. M. Barnes. Zinc deficiency and a synthetic diet for lactose intolerance. Lancet 1:676-677, 1973.

1087. Mueller, C. W., and R. I. Henkin. Determination of zinc in parotid saliva using flameless atomic absorption. Fed. Proc. 33:700, 1974. (abstract)

1088. Mugwira, L. M., and B. D. Knezek. Navy bean responses to zinc fertilizers. Commun. Soil Sci. Plant Anal. 2:337-343, 1971.

1089. Mukherjee, K. L. Microelement composition of sugarcane leaves during their growth and senescence. J. Indian Bot. Soc. 48:180-184, 1969.

1090. Mulay, I. L., R. Roy, B. E. Knox, N. H. Suhr, and W. E. Delaney. Trace-metal analysis of cancerous and noncancerous human tissues. J. Nat. Cancer Inst. 47:1-13, 1971.

1091. Müller, G. Wirkung von Zink bei ungehemmter Hefe-Aldolase. Hoppe-Seyler's Z. Physiol. Chem. 333:246-251, 1963.

1091a. Müntzing, J., and T. Nilsson. Enzyme activity and distribution in the hyperplastic and cancerous human prostate. Scand. J. Urol. Nephrol. 6:107-111, 1972.

1091b. Munch-Petersen, S. The variations in serum copper in the course of 24 hours. Scand. J. Clin. Lab. Invest. 2:48-52, 1950.

1091c. Müntzing, J., T. Nilsson, and J. Polacek. Zinc and β-glucuronidase in the human prostate. Scand. J. Urol. Nephrol. 8:87-90, 1974.

1092. Murakami, H., H. Mori, and S. Taira. Active centre model on the structure and function of the bleomycin molecule as the prototype of esteratic enzymes. J. Theor. Biol. 42:443-460, 1973.

1093. Murphy, E. W., and B. W. Willis. Problems in developing food composition tables. Zinc as a case study. Interface 4(1):3-4, 1975.

1093a. Murphy, E. W., B. W. Willis, and B. K. Watt. Provisional tables on the zinc content of foods. J. Amer. Diet. Assoc. 66:345-355, 1975.

1094. Murphy, J. V. Intoxication following ingestion of elemental zinc. J.A.M.A. 212:2119-2120, 1970.

1095. Murphy, L. S., and L. M. Walsh. Correction of micronutrient deficiencies with fertilizers, pp. 347-387. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972

1096. Murray, J., and S. Rosenthal. The effect of locally applied zinc and aluminum on healing incised wounds. Surg. Gynecol. Obstet. 126: 1298-1300, 1968.

1097. Murthy, G. K., A. S. Goldin, and J. E. Campbell. Zinc-65 in foods. Science 130:1255-1256, 1959.

1098. Mutch, P., and L. S. Hurley. Effect of zinc deficiency during lactation on postnatal growth and development of rats. J. Nutr. 104:828-842, 1974.

1099. Murthy, L., L. M. Klevay, and H. G. Petering. Interrelationships of zinc and copper nutriture in the rat. J. Nutr. 104:1458-1465, 1974.

1100. Murthy, G. K., and U. S. Rhea. Calcium, copper, iron, lead, manganese, and zinc in evaporated milk, infant products, and human milk. J. Dairy Sci. 54:1001-1005, 1971.

1101. Murthy, G. K., U. S. Rhea, and J. T. Peeler. Copper, iron, manganese, strontium, and zinc content of market milk. J. Dairy Sci. 55: 1666-1674, 1972.

1102. Murthy, G. K., U. Rhea, and J. T. Peeler. Levels of antimony, cadmium, chromium, cobalt, manganese, and zinc in institutional total diets. Environ. Sci. Technol. 5:436-442, 1971.

1102a. Mutch, P., and L. S. Hurley. Effect of zinc deficiency during lactation on postnatal growth and development of rats. J. Nutr. 104:828-842, 1974.

1102b. Myers, M. B., and G. Cherry. Zinc and the healing of chronic leg ulcers. Amer. J. Surg. 120:77-81, 1970.

1103. Naji, K., O. Farhri, and K. M. Mousawy. Protective immunity to hydatid disease? Lancet 1:1518-1519, 1973.

1104. Nakatani, R. E. Biological response of rainbow trout (Salmo gairdneri) after ingesting zinc-65, pp. 809-823. In Disposal of Radioactive Wastes into Seas, Oceans, and Surface Waters. Proceedings of a Symposium, Vienna 16-20 May 1966. Vienna: International Atomic Energy Agency, 1966.

1104a. Nandi, D. L., and D. Shemin. δ-Aminolevulinic acid dehydratase of Rhodopseudomonas spheroides. III. Mechanism of porphobilinogen synthesis. J. Biol. Chem. 243:1236-1242, 1968.

1105. Nason, A. Effect of zinc deficiency on the synthesis of tryptophan by Neurospora extracts. Science 112:111-112, 1950.

1106. Nason, A., N. O. Kaplan, and S. P. Colowick. Changes in enzymatic constitution in zinc-deficient Neurospora. J. Biol. Chem. 188:397-406, 1951.

1107. Nassi, L., G. Poggini, C. Vecchi, and P. Galvan. La spettrofotometria ad assorbimento atomico applicata al dosaggio dello zinco "totale", "libero" e "legato" nel colostro e nel latte umano. Boll. Soc. Ital. Biol. Sper. 48:86-89, 1972.

1108. National Research Council. Agricultural Board, Committee on Animal Nutrition, Subcommittee on Dairy Cattle Nutrition. Nutrient Requirements of Domestic Animals. No. 3. Nutrient Requirements of Dairy Cattle. (4th ed.) Washington, D. C.: National Academy of Sciences, 1971. pp. 11, 29.

1109. National Research Council. Agricultural Board, Committee on Animal Nutrition, Subcommittee on Dog Nutrition. Nutrient Requirements of Domestic Animals. No. 8. Nutrient Requirements of Dogs. (Revised 1972) Washington, D. C.: National Academy of Sciences, 1972. pp. 14, 36.

1110. National Research Council, Agricultural Board, Committee on Animal Nutrition, Subcommittee on Fish Nutrition. Nutrient Requirements of Domestic Animals No. 11. Nutrient Requirements of Trout, Salmon, and Catfish. Washington, D. C.: National Academy of Sciences, 1973. 57 pp.

1111. National Research Council. Agricultural Board. Committee on Animal Nutrition, Subcommittee on Horse Nutrition. Nutrient Requirements of Domestic Animals. No. 6. Nutrient Requirements of Horses. (3rd ed.) Washington, D. C.: National Academy of Sciences, 1973. pp. 10-11, 27.

1112. National Research Council. Agricultural Board, Committee on Animal Nutrition, Subcommittee on Laboratory Animal Nutrition. Nutrient Requirements of Domestic Animals. No. 10. Nutrient Requirements of Laboratory Animals. (2nd ed.). Washington, D. C.: National Academy of Sciences, 1972. pp. 33, 48, 53, 64, 72-73.

1113. National Research Council. Agricultural Board. Committee on Animal Nutrition, Subcommittee on Poultry Nutrition. Zinc, p. 11, and Table of nutritional requirements, p. 17. In Nutrient Requirements of Domestic Animals. No. 1. Nutrient Requirements of Poultry. (6th rev. ed.) Washington, D. C.: National Academy of Sciences, 1971.

1114. National Research Council. Agricultural Board, Committee on Animal Nutrition, Subcommittee on Swine Nutrition. Nutrient Requirements of Domestic Animals. No. 2. Nutrient Requirements of Swine. (7th ed.) Washington, D. C.: National Academy of Sciences, 1973. pp. 10-12.

1114a. National Research Council. Agricultural Board. Committee on Animal Nutrition. Subcommittee on Beef Cattle Nutrition. Nutrient Requirements of Domestic Animals No. 4. Nutrient Requirements of Beef Cattle. (4th rev. ed.) Washington, D. C.: National Academy of Sciences, 1970. pp. 10-11, 27.

1115. National Research Council, Agricultural Board and Canadian Department of Agriculture. Atlas of Nutritional Data on United States and Canadian Feeds. Washington, D. C.: National Academy of Sciences, 1971. 772 pp.

1116. National Research Council. Considerations on the Disposal of Radioactive Wastes from Nuclear-Powered Ships into the Marine Environment. NAS Publ. 658. Washington, D.C.: National Academy of Sciences, 1959. 52 pp.

1116a. National Academy of Sciences. National Academy of Engineering. Environmental Studies Board. Water Quality Criteria 1972. A Report of the Committee on Water Quality Criteria. Washington, D. C.: U. S. Government Printing Office, 1974. 594 pp.

1117. National Research Council. Food and Nutrition Board. Proposed Fortification Policy for Cereal-Grain Products. Washington, D. C.: National Academy of Sciences, 1974. 36 pp.

1118. National Research Council. Food and Nutrition Board. Recommended Dietary Allowances. (7th ed.) Washington, D. C.: National Academy of Sciences, 1968. 101 pp.

1119. National Research Council. Food and Nutrition Board. Zinc, pp. 99-101. In Recommended Dietary Allowances. (8th ed.) Washington, D. C.: National Academy of Sciences, 1974.

1119a. National Research Council. Food and Nutrition Board. Committee on Food Protection. Toxicants Occurring Naturally in Foods. (2nd ed.) Washington, D. C.: National Academy of Sciences, 1973. 624 pp.

1120. National Research Council. Food and Nutrition Board. Zinc in Human Nutrition. Summary of Proceedings of a Workshop, Dec. 4-5, 1970. Washington, D. C.: National Academy of Sciences, 1971. 50 pp.

1120a. Natusch, D. S. F., J. R. Wallace, and C. A. Evans, Jr. Toxic trace elements: Preferential concentration in respirable particles. Science 183:202-204, 1974.

1121. Navrot, J., B. Jacoby, and S. Ravikovitch. Fixation of Zn^{65} in some calcareous soils and its availability to tomato plants. Plant Soil 27:141-147, 1967.

1122. Nayler, W. G., and J. E. Anderson. Effects of zinc on cardiac muscle contraction. Amer. J. Physiol. 209:17-21, 1965.

1122a. Neathery, M. W., J. W. Lassiter, R. P. Gentry, and W. J. Miller. Absorption and tissue distribution in rats of zinc-65 grown into young forages versus the inorganic form. J. Dairy Sci. 57:624, 1974. (abstract)

1123. Neathery, M. W., W. J. Miller, D. M. Blackmon, and R. P. Gentry. Performance and milk zinc from low-zinc intake in Holstein cows. J. Dairy Sci. 56:212-217, 1973.

1124. Neathery, M. W., W. J. Miller, D. M. Blackmon, and R. P. Gentry. Zinc-65 metabolism, secretion into milk, and biological half-life in lactating cows. J. Dairy Sci. 56:1526-1530, 1973.

1125. Neathery, M. W., W. J. Miller, D. M. Blackmon, and R. P. Gentry. Zn tissue distribution in lactating Holstein cows fed normal and low-zinc practical diets. J. Anim. Sci. 38:854-859, 1974.

1126. Neathery, M. W., W. P. Miller, D. M. Blackmon, R. P. Gentry, and J. B. Jones. Absorption and tissue zinc content in lactating dairy cows as affected by low dietary zinc. J. Anim. Sci. 37:848-852, 1973.

1127. Neathery, M. W., W. J. Miller, D. M. Blackmon, F. M. Pate, and R. P. Gentry. Effects of long term zinc deficiency on feed utilization, reproductive characteristics, and hair growth in the sexually mature male goat. J. Dairy Sci. 56:98-105, 1972.

1128. Neathery, M. W., S. Rachmat, W. J. Miller, R. P. Gentry, and D. M. Blackmon. Effect of chemical form of oral ^{65}Zn on absorption and metabolism in cattle. Proc. Soc. Exp. Biol. Med. 139:953-956, 1972.

1129. Nelbach, M. E., V. P. Pigiet, Jr., J. C. Gerhart, and H. K. Schachman. A role for zinc in the quaternary structure of aspartate transcarbamylase from Escherichia coli. Biochemistry 11:315-327, 1972.

1129a. Neldner, K. H., and K. M. Hambidge. Zinc therapy of acrodermatitis enteropathica. New Engl. J. Med. 292:879-882, 1975.

1130. Nelson, E. B., A. Cenedella, and N. E. Tolbert. Carbonic anhydrase levels in Chlamydomonas. Phytochemistry 8:2305-2306, 1969.

1131. Nelson, K. W. Nonferrous metallurgical operations, pp. 171-190. In A. C. Stern, Ed. Air Pollution. Vol. 3. Sources of Air Pollution and Their Control. (2nd ed.) New York: Academic Press, 1968.

1132. Nelson, T. S., L. W. Ferrara, and N. L. Storer. Phytate phosphorus content of feed ingredients derived from plants. Poult. Sci. 47:1372-1374, 1968.

1133. Netsky, M. G., W. W. Harrison, M. Brown, and C. Benson. Tissue zinc and human disease. Relation of zinc content of kidney, liver and lung to atherosclerosis and hypertension. Amer. J. Clin. Path. 51:358-365, 1969.

1134. Neuberger, A., and G. H. Tait. Studies on the biosynthesis of porphyrin and bacteriochlorophyll by Rhodopseudomonas spheroides. V. Zinc-protoporphyrin chelatase. Biochem. J. 90:607-616, 1964.

1135. Neubert, P., W. Wrazidlo, H. P. Vielemeyer, I. Hundt, F. Gullmick, and W. Bergmann. Research Report, Institut für Pflanzenernahrung, Jena, Berlin, 1969.

1136. Wilfong, R. F., and D. M. Neville, Jr. The isolation of a brush border membrane fraction from rat kidney. J. Biol. Chem. 245:6106-6112, 1970.

1137. Newton, D., and A. Holmes. A case of accidental inhalation of zinc-65 and silver-110m. Radiat. Res. 29:403-412, 1966.

1138. Niebrój, T. K. On the cytochemical localization of zinc in the full-term placenta in normal and pathological conditions. Folia Histochem. Cytochem. 5:163-174, 1967.

1139. Nielsen, F. H., R. P. Dowdy, and Z. Z. Ziporin. Effect of zinc deficiency on sulfur-35 and hexosamine metabolism in the epiphyseal plate and primary spongiosa of the chick. J. Nutr. 100:903-908, 1970.

1139a. Nielsen, F. H., M. L. Sunde, and W. G. Hoekstra. Effect of histamine, histidine, and some related compounds on the zinc-deficient chick. Proc. Soc. Exp. Biol. Med. 124:1106-1110, 1967.

1140. Nielsen, F. H., M. L. Sunde, and W. G. Hoekstra. Effect of dietary amino acid source on the zinc-deficiency syndrome in the chick. J. Nutr. 89:24-34, 1966.

1141. Nielsen, F. H., M. L. Sunde, and W. G. Hoekstra. Effect of some dietary synthetic and natural chelating agents on the zinc-deficiency syndrome in the chick. J. Nutr. 89:35-42, 1966.

1142. Nielsen, F. H., and Z. Z. Ziporin. Effect of zinc deficiency on the uptake of $^{35}SO_4^=$ by the epiphyseal plate and primary spongiosa of the chick. Fed. Proc. 28:762, 1969. (abstract)

1143. Niklowitz, W. J., and D. W. Yeager. Interference of Pb with essential brain tissue Cu, Fe, and Zn as main determinant in experimental tetraethyllead encephalopathy. Life Sci. 13:897-905, 1973.

1143a. Nilsson, T., E. Schueller, and W. Staubitz. β-glucuronidase activity of the epithelial cells and stroma cells in prostatic hyperplasia. Invest. Urol. 11:145-148, 1973.

1144. Nixon, G. S., H. D. Livingston, and H. Smith. Estimation of zinc in human enamel by activation analysis. Arch. Oral Biol. 12:411-416, 1967.

1144a. Norman, M., S. Gamulin, and K. Clark. The distribution of ribosomes between different functional states in livers of fed and starved mice. Biochem. J. 134:387-398, 1973.

1144b. Nordberg, G. F., Ed. Effects and Dose-Response Relationships of Toxic Metals. Proceedings from an International Meeting, Tokyo, Nov. 18-23, 1974. New York: Elsevier Scientific Publishing Company, 1976. 559 pp.

1145. Noro, L., and U. Uotila. Zinc stearate lund, p. 26. In Atti XI Congresso Internazionale de Medicina del Lavoro, Napoli 13-19 Settembre 1954. Vol. 3. Napoli: Istituto di Medicina del Lavoro, 1954.

1146. Norrdin, R. W., L. Krook, W. G. Pond, and E. F. Walker. Experimental zinc deficiency in weanling pigs on high and low calcium diets. Cornell Vet. 63:264-290, 1973.

1147. Northrop, D. B. Transcarboxylase. VI. Kinetic analysis of the reaction mechanism. J. Biol. Chem. 244:5808-5819, 1969.

1148. Northrop. D. B., and H. G. Wood. Transcarboxylase. V. The presence of bound zinc and cobalt. J. Biol. Chem. 244:5801-5807, 1969.

1149. Norvell, W. A., and W. L. Lindsay. Reactions of DTPA chelates of iron, zinc, copper, and manganese with soils. Soil Sci. Soc. Amer. Proc. 36:778-783, 1972.

1150. Nougues, C., and M. Lamand. Possibilities et limites de l'utilisation du poil dans le diagnostic de la carence en zinc chez le bovin. Ann. Rech. Vet. 3:505-509, 1972.

1151. Nyman, P. O. Purification and properties of carbonic anhydrase from human erythrocytes. Biochim. Biophys. Acta 52:1-12, 1961.

1152. Oberleas, D., and A. S. Prasad. Growth as affected by zinc and protein nutrition. Amer. J. Clin. Nutr. 22:1304-1314, 1969.

1153. Oberleas, D., and A. S. Prasad. The dynamics of nucleic acid synthesis in zinc deficiency. Fed. Proc. 33:699, 1974. (abstract)

1154. Oberleas, D., J. K. Seymour, R. Lenaghan, J. Hovanesian, R. F. Wilson, and A. S. Prasad. Effect of zinc deficiency on wound-healing in rats. Amer. J. Surg. 121:566-568, 1971.

1155. O'Dell, B. L. Effect of dietary components upon zinc availability. A review with original data. Amer. J. Clin. Nutr. 22:1315-1322, 1969.

1156. O'Dell, B. L., C. E. Burpo, and J. E. Savage. Evaluation of zinc availability in foodstuffs of plant and animal origin. J. Nutr. 102:653-660, 1972.

1157. O'Dell, B. L., and J. E. Savage. Effect of phytic acid on zinc availability. Proc. Soc. Exp. Biol. Med. 103:304-306, 1960.

1158. Oden, S., B. Berggren, and A. G. Engvall. Heavy metals and chlorinated hydrocarbons in sludge. Grundforbättring 23(Specialnummer 5):55-68, 1970. (in Swedish)

1159. Odum, E. P. Excretion rate of radio-isotopes as indices of metabolic rates in nature: Biological half-life of zinc-65 in relation to temperature, food consumption, growth and reproduction in arthropods. Biol. Bull. 121:371-372, 1961. (abstract)

1160. Odum, E. P., and F. B. Golley. Radioactive tracers as an aid to the measurement of energy flow at the population level in nature, pp. 403-410. In V. Schultz and A. W. Klement, Jr., Eds. Radioecology. New York: Reinhold, 1963.

1161. Ohaniance, L., and P. Chaix. Effect inhibiteur de Zn^{2+} sur la biosynthese induite par l'oxygene des enzymes respiratoires de la levure. Biochim. Biophys. Acta 128:228-238, 1966.

1162. Oiwa, K., T. Kimura, H. Makino, and M. Okuda. Determination of zinc in blood serum and red cells by atomic absorption spectrophotometry. Bunseki Kagaku (Jap. Anal.) 17:810-815, 1968. (in Japanese)

1163. O'Kelley, J. C. Mineral nutrition of algae. Ann. Rev. Plant Physiol. 19: 89-112, 1968.

1163a. Okereke, T., I. Sternlieb, A. G. Morell, and I. H. Scheinberg. Systemic absorption of intrauterine copper. Science 177:358-360, 1972.

1164. Okunewick, J. P., S. E. Herrick, and T. G. Hennessy. Effects of plasma on the absorption of zinc by mammalian erythrocytes. J. Cell. Comp. Physiol. 63:333-337, 1964.

1164a. O'Leary, J. A., and W. N. Spellacy. Zinc and copper levels in pregnant women and those taking oral contraceptives. Amer. J. Obstet. Gynecol. 103:131-132, 1969.

1165. Olehy, D. A., R. A. Schmitt, and W. F. Bethard. Neutron activiation analysis of magnesium, calcium, strontium, barium, manganese, cobalt, copper, zinc, sodium, and potassium in human erythrocytes and plasma. J. Nucl. Med. 7:917-927, 1966.

1166. Oldendorf, W. H. Brain uptake of radiolabeled amino acids, amines, and hexoses after arterial injection. Amer. J. Physiol. 221:1629-1639, 1971.

1167. Oliver, A. R. Effluent treatment processes and their economics. Metal Finish. J. 18:316-322, 327, 1972.

1168. Olsen, S. R. Micronutrient interactions, pp. 243-264. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

1169. Olsen, S. R., and W. D. Kemper. Movement of nutrients to plant roots. Adv. Agron. 20:91-151, 1968.

1170. Olson, A. D., and W. B. Hamlin. Serum copper and zinc by atomic absorption spectrophotometry. Atom. Absorpt. Newslett. 7:69-71, 1968.

1171. Olson, K. B., G. Heggen, C. F. Edwards, and L. W. Gorham. Trace element content of cancerous and noncancerous human liver tissue. Science 119:772-773, 1954.

1171a. Olsson, R. A., and H. E. Ticktin. Zinc metabolism in acute intermittent porphyria. J. Lab. Clin. Med. 60:48-52, 1962.

1172. O'Neal, R. M., G. W. Pla, M. R. S. Fox, F. S. Gibson, and B. E. Fry, Jr. Effect of zinc deficiency and restricted feeding on protein and ribonucleic acid metabolism of rat brain. J. Nutr. 100:491-497, 1970.

1172a. Ondov, J. M., W. H. Zoller, and G. E. Gordon. Trace elements on aerosols from motor vehicles. Paper 74-197 Presented at the 67th Annual Meeting of the Air Pollution Control Association, Denver, Colorado, June 9-13, 1974. 24 pp.

1173. Oppenheimer, H. L., R. W. Green, and R. H. McKay. Function of zinc in horse liver alcohol dehydrogenase. Arch. Biochem. Biophys. 119: 552-559, 1967.

1174. Orgebin-Crist, M.-C., M. Freeman, and G. H. Barney. Sperm formation in zinc-deficient rats. Ann. Biol. Anim. Biochem. Biophys. 11: 547-558, 1971.

1175. Orinchak, M. A. Zinc content in infant nutrition products and index of alimentary zinc provision for healthy infants. Mikroelem. Med. 5: 131-133, 1974. (in Russian)

1176. Deleted.

1177. O'Rourke, J., J. Durrani, C. Benson, J. Bronzino, and C. Miller. Studies in uveal physiology. III. Anterior chamber clearance, uveoretinal distribution, and respiratory response associated with zinc 69m. Arch. Ophthalmol. 88:185-192, 1972.

1178. Orten, J. M. Biochemical aspects of zinc metabolism, pp. 38-46. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1179. Osis, D., L. Kramer, E. Wiatrowski, and H. Spencer. Dietary zinc intake in man. Amer. J. Clin. Nutr. 25:582-588, 1972.

1180. Osmanski, C. P., and J. Meyer. Ultrastructural changes in buccal and palatal mucosa of zinc deficient rats. J. Invest. Derm. 53:14-28, 1969.

1181. Osterberg, C., L. D. Kulm, and J. V. Byrne. Gamma emitters in marine sediments near the Columbia River. Science 139:916-917, 1963.

1182. Osterberg, C., J. Pattullo, and W. Pearcy. Zinc 65 in euphasiids as related to Columbia River water off the Oregon coast. Limnol. Oceanogr. 9:249-257, 1964.

1183. Osterberg, C., L. Small, and L. Hubbard. Radioactivity in large marine plankton as a function of surface area. Nature 197:883-884, 1963.

1184. Ottolenghi, A. C. Phospholipase C from Bacillus cereus, a zinc-requiring metalloenzyme. Biochim. Biophys. Acta 106:510-518, 1965.

1185. Ou, J. T. Effect of Zn^{2+} on bacterial conjugation: Increase in ability of F-cells to form mating pairs. J. Bacteriol. 115:648-654, 1973.

1186. Ou, J. T., and T. F. Anderson. Effect of Zn^{2+} on the adsorption of male-specific filamentous deoxyribonucleic acid and isometric ribonucleic acid bacteriophages. J. Virol. 10:869-871, 1972.

1186a. Ovadia, J., J. W. McArthur, L. Kopito, and H. Ulfelder. The cervical mucus secretion of the Bonnet monkey (M. radiata): Anatomical basis and physiological regulation. Biol. Reprod. 5:127-145, 1971.

1187. Owen, A. A., E. R. Peo, Jr., P. J. Cunningham, and B. D. Moser. Chelated trace minerals for G-F swine. J. Anim. Sci. 37:95-103, 1973.

1188. Owen, A. A., E. R. Peo, Jr., P. J. Cunningham, and B. D. Moser. Effect of EDTA on utilization of dietary zinc by G-F swine. J. Anim. Sci. 37:470-478, 1973.

1189. Oxford, J. S., and D. D. Perrin. Inhibition of the particle-associated RNA-dependent RNA polymerase activity of influenza viruses by chelating agents. J. Gen. Virol. 23:59-71, 1974.

1190. Ozanne, P. G. The effect of nitrogen on zinc deficiency in subterranean clover. Austral. J. Biol. Sci. 8:47-55, 1955.

1191. Oziebło, L. Estimation of zinc and cobalt content in human semen. Endokrynol. Pol. 24:91-95, 1973. (in Polish, summary in English)

1191a. Page, A. L. Fate and Effects of Trace Elements in Sewage Sludge When Applied to Agricultural Lands. A Literature Review. EPA-670/2-74-005. Riverside: University of California, Department of Soil Science and Agricultural Engineering, 1974. 96 pp.

1191b. Painter, H. A. Chemical, physical, and biological characteristics of wastes and waste effluents, pp. 329-364. In L. L. Ciaccio, Ed. Water and Water Pollution Handbook. Vol. 1. New York: Marcel Dekker, Inc., 1971.

1192. Pak, C. Y. C. Sodium cellulose phosphate: Necrosis of action and effect on mineral metabolism. J. Clin. Pharm. 13:15-27, 1973.

1193. Pak, C. Y. C., B. Ruskin, and E. Diller. Enhancement of renal excretion of zinc by hydrochlorothiazide. Clin. Chim. Acta 39:511-517, 1972.

1194. Pallauf, J., and M. Kirchgessner. Effekt verschiedener Mn-bzw. Cu-Zulagen bei mangelnder Zinkversorgung. Zentralbl. Vetinaermed. (A) 21:562-571, 1974.

1195. Pallauf, J., and M. Kirchgessner. Experimenteller Zinkmangel bei wachsenden Ratten. 2. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 28:128-139, 1971.

1196. Pallauf, J., and M. Kirchgessner. Konzentration und Verteilung des Zinks im Organismus. 1. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 28:121-128, 1971.

1197. Pallauf, J., and M. Kirchgessner. Zinkgehalte in Knochen und Ganzkörper wachsender Ratten bei unterschiedlicher Zinversorgung. 5. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 30:193-202, 1972.

1198. Pallauf, J., and M. Kirchgessner. Zinkkonzentration des Rattenhaares bei Zink-depletion und-repletion. Zentralbl. Vet. Med. A 20:100-109, 1973.

1199. Pallauf, J., and M. Kirchgessner. Zinkkonzentration in Blut und Serum wachsender Ratten bei zinkmangel. Zentralbl. Vet. Med. A 19:594-604, 1972.

1200. Pallauf, J., and M. Kirchgessner. Zur Wirksamkeit erhöhter Zulagen an Vitamin B_1, B_2, B_6, B_{12}, Pantothen- und Nicotinsäure bei Zinkmangel. Int. Z. Vit. Ernährungsforsch. 43:339-350, 1973.

1201. Pallauf, J., and M. Kirchgessner. Zur Wirksamkeit von Biotin- und Folsäurezulagen bei Zinkmangel. Int. Z. Vit. Ernährungsforsch. 42:555-564, 1972.

1202. Palludan, B., and I. Wegger. Zinc metabolism in swine. III. Placental transfer of zinc in normal and zinc deficient gilts and its influence on foetal development. Aarsberetn. Inst. Sterilitetsforsk. Kgl. Vet. Landbohoejsk. 15:27-53, 1972. (in Danish, summary in English)

1203. Panchiznaya, E. S. The content of zinc in the venous and menstrual blood of healthy women. Akush. Ginekol. (Mosk.) 43(1):37-39, 1967. (in Russian)

1203a. Parashchak, A. P. Iron, copper, cobalt and zinc metabolism in a cinchophen gastric ulcer in the dog. Biol. Nauki 3:36-42, 1972. (in Russian)

1204. Papasteriade, A. A. Study of Conditions Due to Low Levels of Zinc in Sheep and Goats in Greece. Ph.D. Thesis. Thessalonika: Aristotelian University of Thessalonika, 1973. pp. 177-442. (in Greek)

1205. Parisi, A. F., and B. L. Vallee. Isolation of a zinc α_2-macroglobulin from human serum. Biochemistry 9:2421-2426, 1970.

1206. Pařízek, J. The destructive effect of cadmium ion on testicular tissue and its prevention by zinc. J. Endocrinol. 15:56-63, 1957.

1206a. Parizek, J., and Z. Zahor. Effect of cadmium salts on testicular tissue. Nature 177:1036, 1956.

1207. Parkash, S., and R. Jenness. Status of zinc in cow's milk. J. Dairy Sci. 50:127-134, 1967.

1208. Parker, M. M., F. L. Humoller, and D. J. Mahler. Determination of copper and zinc in biological material. Clin. Chem. 13:40-48, 1967.

1209. Parker, P. L. Zinc in a Texas Bay. Publ. Inst. Marine Sci. Univ. Texas 8:75-79, 1962.

1210. Parr, R. M., and D. M. Taylor. The concentrations of cobalt, copper, iron and zinc in some normal human tissues as determined by neutron-activation analysis. Biochem. J. 91:424-431, 1964.

1211. Partington, J. R. [Brass, zinc, copper and bronze], pp. 83, 475. In Origins and Development of Applied Chemistry. New York: Longmans, Green and Co., 1935.

1211a. Parzyck, D. C. Toxicity of Cadmium in Pregnant Rats Fed a Zinc-Deficient Diet. Ph.D. Thesis. Lafayette, Ind.: Purdue University, 1974. 184 pp.

1212. Pate, F. M., W. J. Miller, D. M. Blackmon, and R. P. Gentry. Early tissue ^{65}Zn distribution after duodenal dosing in calves fed zinc-deficient and control diets. Proc. Soc. Exp. Biol. Med. 135:653-656, 1970.

1213. Pate, F. M., W. J. Miller, D. M. Blackmon, and R. P. Gentry. $^{65}Zinc$ absorption rate following single duodenal dosing in calves fed zinc-deficient or control diets. J. Nutr. 100:1259-1266, 1970.

1213a. Patel, H. M., and B. E. Ryman. α-mannosidase in zinc-deficient rats: Possibility of liposomal therapy in mannosidosis. Biochem. Soc. Trans. 2:1014-1017, 1974.

1213b. Patek, A. J., Jr., and C. Haig. The occurrence of abnormal dark adaptation and its relation to vitamin A metabolism in patients with cirrhosis of the liver. J. Clin. Invest. 18:609-616, 1939.

1214. Pauley, G. B., and R. E. Nakatani. Metabolism of the radioisotope ^{65}Zn in the freshwater mussel Anodonta californiensis. J. Fish. Res. Board Can. 25:2691-2694, 1968.

1215. Pearcy, W. C., and C. L. Osterberg. Zinc-65 and manganese-54 in albacore Thunnus alalunga from the west coast of North America. Limnol. Oceanogr. 13:490-498, 1968.

1216. Pearcy, W. G., and H. A. Vanderploeg. Radioecology of benthic fishes off Oregon, pp. 245-261. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA in Seattle U.S.A. 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

1216a. Pécoud, A., P. Donzel, and J. L. Schelling. Effect of foodstuffs on the absorption of zinc sulfate. Clin. Pharmacol. Ther. 17:469-474, 1975.

1217. Peifer, J. J., and L. L. Crawford. Serum, urine and hair zinc levels of preadolescents from black and white, middle and low income families. Fed. Proc. 34:896, 1975. (abstract)

1217a. Pekarek, R. S., and W. R. Beisel. Effect of endotoxin on serum zinc concentrations in the rat. Appl. Microbiol. 18:482-484, 1969.

1217b. Pekarek, R. S., and W. R. Beisel. Charaterization of the endogenous mediator(s) of serum zinc and iron depression during infection and other stresses. Proc. Soc. Exp. Biol. Med. 138:728-732, 1971.

1218. Pekarek, R. S., and M. C. Powanda. Protein synthesis in zinc deficient rats during tularemia. Fed. Proc. 34:882, 1975. (abstract)

1219. Pekarek, R. S., R. W. Wannemacher, Jr., and W. R. Beisel. The effect of leukocytic endogenous mediator (LEM) on the tissue distribution of zinc and iron. Proc. Soc. Exp. Biol. Med. 140:685-688, 1972.

1219a. Pekarek, R. S., M. C. Powanda, and R. W. Wannemacher, Jr. The effect of leukocytic endogenous mediator (LEM) on serum copper and ceruloplasmin concentrations in the rat. Proc. Soc. Exp. Biol. Med. 141:1029-1031, 1972.

1220. Pekas, J. C. Zinc 65 metabolism: Gastrointestinal secretion by the pig. Amer. J. Physiol. 211:407-413, 1966.

1221. Penhos, J. C., E. D. Brown, J. Hsu, and J. C. Smith, Jr. Oral glucose tolerance in zinc deficient rats. Fed. Proc. 33:700, 1974. (abstract)

1222. Pentreath, R. J. The accumulation from water of ^{65}Zn, ^{54}Mn, ^{58}Co, and ^{59}Fe by the mussel, Mytilus edulis. J. Marine Biol. Assoc. U. K. 53:127-143, 1973.

1223. Pentreath, R. J. The metabolism of radionuclides, pp. 97-126. In Marine Radioecology: The Cycling of Artificial Radionuclides Through Marine Food Chains. (Second European Nuclear Energy Agency Seminar on Marine Radioecology, Hamburg, 20th-24th September, 1971. Paris: European Nuclear Energy Agency, 1971.

1224. Pequegnat, J. E., S. W. Fowler, and L. F. Small. Estimates of the zinc requirements of marine organisms. J. Fish. Res. Board Can. 26:145-150, 1969.

1225. Perhac, R. M. Distribution of Cd, Co, Cu, Fe, Mn, Ni, Pb, and Zn in dissolved and particulate solids from two streams in Tennessee. J. Hydrol. 15:177-186, 1972.

1226. Perkins, D. J. Zn^{2+} binding to poly-L-glutamic acid and human serum albumin. Biochim. Biophys. Acta 86:635-636, 1964.

1227. Perkins, R. W., J. L. Nelson, and W. L. Haushild. Behavior and transport of radionuclides in the Columbia River between Hanford and Vancouver, Washington. Limnol. Oceanogr. 11:235-248, 1966.

1228. Perkins, R. W., and J. M. Nielsen. Zinc-65 in foods and people. Science 129:94-95, 1959.

1229. Perkins, R. W., J. M. Nielsen, W. C. Roesch, and R. C. McCall. Zinc-65 and chromium-51 in foods and people. Science 132:1895-1897, 1960.

1230. Pernis, B., E. C. Vigliani, G. Caragna, and M. Funilli. Endogenous pyrogen in the pathogenesis of zinc-fume fever. Med. Lavoro 51:579-586, 1960.

1231. Perrin, D. D., and V. S. Sharma. The stability constants of metal-adenosine triphosphate complexes. Biochim. Biophys. Acta 127:35-41, 1966.

1232. Perry, H. M., Jr., I. H. Tipton, H. A. Schroeder, and M. J. Cook. Variability in the metal content of human organs. J. Lab. Clin. Med. 60:245-253, 1962.

1232a. Perry, H. O. Acrodermatitis enteropathica, Unit 1-8. In D. J. Demis, R. G. Crounse, R. L. Dobson, and J. McGuire, Eds. Clinical Dermatology. Vol. 1. New York: Harper & Row, Publishers, 1975.

1232b. Perry, H. M., Jr., and H. A. Schroeder. Lesions resembling vitamin B complex deficiency and urinary loss of zinc produced by ethylenediamine tetraacetate. Amer. J. Med. 22:168-172, 1957.

1233. Perry, R. Mass spectrometry in the detection and identification of air pollutants, pp. 130-137. In B. Westley, Ed. Proceedings of International Symposium on Identification and Measurement of Environmental Pollutants. Ottawa, Ontario, Canada, June 14-17, 1971. Ottawa: National Research Council of Canada, 1971.

1234. Perry, R. H., C. H. Chilton, S. D. Kirkpatrick, Eds. Chemical Engineers' Handbook. (4th ed.) New York: McGraw-Hill Book Company, 1963.

1235. Persigehl, M., A. Höck, K. Kasperek, E. Land, and L. E. Feinendegen. Änderung der Zinkkonzentration im Serum bei verschiedenen Stoffwechselsituationen. Z. Klin. Chem. Klin. Biochem. 12:171-175, 1974.

1235a. Petering, H. G., H. H. Buskirk, and J. A. Crim. Effect of dietary mineral supplements of the rat on the antitumor activity of 3-ethoxy-2-oxobutyraldehyde bis(thiosemicarbazone). Cancer Res. 27:1115-1121, 1967.

1235b. Petering, H. G., D. W. Yeager, and S. O. Witherup. Trace metal content of hair. I. Zinc and copper content of heman hair in relation to age and sex. Arch. Environ. Health 23:202-207, 1971.

1235c. Peters, H. A. BAL therapy of acute porphyrinuria. Neurology 4:477-479, 1954.

1236. Petering, H. G., M. A. Johnson, and K. L. Stemmer. Studies of zinc metabolism in the rat. I. Dose-response effects of cadmium. Arch. Environ. Health 23:93-101, 1971.

1236a. Peterson, J. R., C. Lue-Hing, and D. R. Zenz. Chemical and biological quality of municipal sludge, pp. 26-37. In W. E. Sopper and L. T. Kardos, Eds. Recycling Treated Municipal Wastewater and Sludge Through Forest and Cropland. Proceedings of a Symposium. University Park: The Pennsylvania State University Press, 1973.

1237. Peterson, P. J. The distribution of zinc-65 in Agrostis tenuis Sibth. and A. stolonifera L. tissues. J. Exp. Bot. 20:863-875, 1969.

1238. Pettigrew, D. W. Annual Review 1971. U. S. Zinc Industry Including Statements from Other Counties. New York: Zinc Institute, 1971. 28 pp.

1239. Pfeiffer, C. C., and V. Iliev. A study of zinc deficiency and copper excess in schizophrenias. Int. Rev. Neurobiol. (Suppl. 1):141-165, 1972.

1240. Pfeiffer, C. C., and E. H. Jenney. Fingernail white spots: Possible zinc deficiency. J.A.M.A. 228:157, 1974.

1241. Pfeilsticker, K. Spurenelemente in Organen und Urin bei Krebs. Z. Klin. Chem. 3:145-150, 1965.

1241a. Phatak, S. S., and V. N. Patwardhan. Toxicity of nickel. J. Sci. Ind. Res. 9b(3):70-76, 1950.

1242. Phillips, J. L., and P. Azari. Effect of iron transferrin on nucleic acid synthesis in phytohemagglutinin-stimulated human lymphocytes. Cell. Immunol. 15:94-99, 1975.

1243. Phillips, J. L., and P. Azari. Zinc transferrin. Enhancement of nucleic acid synthesis in phytohemagglutinin-stimulated human lymphoctyes. Cell. Immunol. 10:31-37, 1974.

1244. Picciano, M. F., and H. A. Guthrie. Determination of concentrations and variations of copper, iron and zinc in human milk. Fed. Proc. 32: 929, 1973. (abstract)

1245. Pickering, Q. H. Some effects of dissolved oxygen concentrations upon the toxicity of zinc to bluegill Lepomis macrochirus Raf. Water Res. 2:187-194, 1968.

1246. Pickering, Q. H., and C. Henderson. The acute toxicity of some heavy metals to different species of warmwater fishes. Int. J. Air Water Pollut. 10:453-463, 1966.

1246a. Piggott, L., D. Caldwell, and D. Oberleas. Zinc deficiency, disturbed children, and civil rights. Biol. Psychiatr. 9:325-327, 1974.

1247. Pillay, K. K. S., and C. C. Thomas, Jr. Determination of the trace element levels in atmospheric pollutants by neutron activation analysis. J. Radioanal. Chem. 7:107-118, 1971.

1248. Piras, R., and B. L. Vallee. Procarboxypeptidase A-carboxypeptidase A interrelationships. Metal and substrate binding. Biochemistry 6: 348-357, 1967.

1249. Piro, A., M. Bernhard, M. Branica, and M. Verzi. Incomplete exchange reaction between radioactive ionic zinc and stable natural zinc in seawater, pp. 29-45. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA in Seattle U.S.A., 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

1249a. Piscator, M. Cadmium-zinc interactions, pp. 951-959. In International Symposium on Recent Advances in the Assessment of the Health Affects of Environmental Pollution, Paris, June 24-28, 1974. Vol. II. 1975.

1250. Piscator, M., and B. Lind. Cadmium, zinc, copper, and lead in human renal cortex. Arch. Environ. Health 24:426-431, 1972.

1251. Pitt, R. E., and G. Amy. Atomic absorption analysis of individual land-use samples, pp. 19-56. In Toxic Materials Analysis of Street Surface Contaminants. EPA-R2-73-283. San Mateo, Calif.: URS Research Co., 1973.

1252. Plantin, L. P., and P. O. Strandberg. Whole blood concentrations of copper and zinc in rheumatoid arthritis studied by activation analysis. Acta Rheumatol. Scand. 11:30-34, 1965.

1253. Platte, J. A., and V. M. Marcy. Photometric determination of zinc with zincon. Application to water containing heavy metals. Anal. Chem. 31:1226-1228, 1959.

1254. Plocke, D. J., C. Levinthal, and B. L. Vallee. Alkaline phosphatase of Escherichia coli: A zinc metalloenzyme. Biochemistry 1:373-378, 1962.

1255. Poiesz, B. J., N. Battula, and L. A. Loeb. Zinc in reverse transcriptase. Biochem. Biophys. Res. Commun. 56:959-964, 1974.

1256. Poiesz, B. J., G. Seal, and L. A. Loeb. Reverse transcriptase. Correlation of zinc content with activity. Proc. Nat. Acad. Sci. U.S.A. 71:4892-4896, 1974.

1257. Polikarpov, G. G. Radioecology of Aquatic Organisms. (English translation edited by V. Schultz, and A. W. Klement, Jr.) New York: Reinhold Book Division, 1966. (translation from Russian) 314 pp.

1258. Polson, E. E., and M. W. Adams. Differential response of navy beans (Phaseolus vulgaris L.) to zinc. I. Differential growth and elemental composition at excessive Zn levels. Agron. J. 62:557-560, 1970.

1259. Pond, W. G., P. Chapman, and E. Walker, Jr. Influence of dietary zinc, corn oil and cadmium on certain blood components, weight gain and parakeratosis in young pigs. J. Anim. Sci. 25:122-127, 1966.

1260. Poortmans, J. R., and K. Schmid. The level of Zn-α_2-glycoprotein in normal human body fluids and kidney extract. J. Lab. Clin. Med. 71: 807-811, 1968.

1261. Pories, W. J., and W. H. Strain. Zinc sulfate therapy in surgical patients, pp. 139-157. In W. J. Pories, W. H. Strain, J. M. Hsu, and R. L. Woosley, Eds. Clinical Applications of Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1974.

1262. Pories, W. J., J. H. Henzel, C. G. Rob, and W. H. Strain. Acceleration of healing with zinc sulfate. Ann. Surg. 165:432-436, 1967.

1263. Pories, W. J., J. M. Henzel, C. G. Rob, and W. H. Strain. Acceleration of wound healing in man with zinc sulphate given by mouth. Lancet 1:121-124, 1967.

1264. Pories, W. J., and W. H. Strain. The functional role of zinc in epidermal tissues, pp. 75-77. In C. F. Mills, Ed. Trace Element Metabolism in Animals. Proceedings of WAAP/IBP International Symposium, Aberdeen, Scotland, July 1969. London: E. & S. Livingston, 1970.

1265. Pories, W. J., and W. H. Strain. Zinc and wound healing, pp. 378-391. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1266. Portmann, J. E. The levels of certain metals in fish from coastal waters around England and Wales. Aquaculture 1:91-96, 1972.

1266a. Portnoy, B., and M. Molokhia. Zinc in acrodermatitis enteropathica. Lancet 2:663-664, 1974. (letter)

1267. Possingham, J. V. The effect of mineral nutrition on the content of free amino acids and amides in tomato plants. I. A comparison of the effects of deficiencies of copper, zinc, manganese, iron, and molybdenum. Austral. J. Biol. Sci. 9:539-551, 1956.

1268. Poswillo, D. E., and B. Cohen. Inhibition of carcinogenesis by dietary zinc. Nature 231:447-448, 1971.

1268a. Pound, C. E., and R. W. Crites. Characteristics of municipal effluents, pp. 49-61. In Proceedings of the Joint Conference on Recycling Municipal Sludges and Effluents on Land. Champaign, Illinois, July 9-13, 1973. Washington, D. C.: National Association of State Universities and Land Grant Colleges, 1973.

1268b. Powanda, M. C., G. L. Cockerell, and R. S. Pekarek. Amino acid and zinc movement in relation to protein synthesis early in inflammation. Amer. J. Physiol. 225:399-401, 1973.

1269. Powell, G. W., W. Y. Miller, J. D. Morton, and C. M. Clifton. Influence of dietary cadmium level and supplemental zinc on cadmium toxicity in the bovine. J. Nutr. 84:205-214, 1964.

1269a. Powell, H. E., H. Fukubayashi, L. W. Higley, and L. L. Smith. Recovery of Zinc, Copper, and Lead-Tin Mixtures from Brass Smelter Flue Dusts. Bureau of Mines Report of Investigations 7637. Washington, D. C.: U. S. Department of the Interior, 1972. 8 pp.

1270. Prahl, J. W., and H. Neurath. Pancreatic enzymes of the spiny Pacific dogfish. II. Procarboxypeptidase B and carboxypeptidase B. Biochemistry 5:4137-4145, 1966.

1271. Praissman, M., and J. A. Rupley. Comparison of protein structure in the crystal and in solution. II. Tritium-hydrogen exchange of zinc-free and zinc insulin. Biochemistry 7:2431-2445, 1968.

1272. Prakash, N. J., J. Fontana, and R. I. Henkin. Effect of transitional metal ions on $(Na^+ + K^+)$ ATPase activity and the uptake of norepinephrine and choline by brain synaptosomes. Life Sci. (Part 1) 12:249-259, 1973.

1272a. Henkin, R. I., R. D. Powell, and N. K. Prakash. Testicular steroidogenesis and morphology in zinc deficiency, Abstract 359, p. A-228. In Endocrine Society Program of the 55th Annual Meeting, Chicago, 1973. Bethesda, Md.: The Endocrine Society, 1973.

1273. Delete 1273--use 1283.

1273a. Prasad, A. S. Metabolism of zinc and its deficiency in human subjects, pp. 250-301. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1274. Prasad, A. S. Nutritional metabolic role of zinc. Fed. Proc. 26:172-185, 1967.

1275. Prasad, A. S., M. Diwany, M. Gabr, H. H. Sandstead, N. Mokhtar, and A. El Hefny. Biochemical studies in thalassemia. Ann. Intern. Med. 62: 87-96, 1965.

1276. Prasad, A. S., J. A. Halsted, and M. Nadimi. Syndrome of iron deficiency anemia, hepatosplenomegaly, hypogonadism, dwarfism and geophagia. Amer. J. Med. 31:532-546, 1961.

1277. Prasad, A. S., A. Miale, Jr., Z. Farid, H. H. Sandstead, and A. R. Schulert. Zinc metabolism in patients with syndrome of iron deficiency anemia, hepatosplenomegaly, dwarfism, and hypogonadism. J. Lab. Clin. Med. 61:537-549, 1963.

1278. Prasad, A. S., A. Miale, Jr., Z. Farid, H. H. Sandstead, A. R. Schulert, and W. J. Darby. Biochemical studies on dwarfism, hypogonadism, and anemia. Arch. Intern. Med. 111:407-428, 1963.

1278a. Prasad, A. S., and D. Oberleas. Binding of zinc to amino acids and serum proteins in vitro. J. Lab. Clin. Med. 76:416-425, 1970.

1279. Prasad, A. S., and D. Oberleas. Changes in activities of zinc-dependent enzymes in zinc-deficient tissues of rats. J. Appl. Physiol. 31: 842-846, 1971.

1280. Prasad, A. S., and D. Oberleas. Thymidine kinase activity and incorporation of thymidine into DNA in zinc-deficient tissue. J. Lab. Clin. Med. 83:634-639, 1974.

1281. Prasad, A. S., and D. Oberleas. Zinc deficiency in man. Lancet 1:463-464, 1974. (letter)

1282. Prasad, A. S., and D. Oberleas. Zinc deficiency in man. Lancet 1:1520-1521, 1973.

1283. Prasad, A. S., D. Oberleas, and J. A. Halsted. Determination of zinc in biological fluids by atomic absorption spectrophotometry in normal and cirrhotic subjects. J. Lab. Clin. Med. 66:508-516, 1965.

1284. Prasad, A. S., D. Oberleas, D. Koniuch, and E. DuMouchelle. Ribonuclease and deoxyribonuclease activities in zinc-deficient tissues. J. Lab. Clin. Med. 82:461-466, 1973.

1285. Prasad, A. S., D. Oberleas, E. R. Miller, and R. W. Luecke. Biochemical effects of zinc deficiency: Changes in activities of zinc-dependent enzymes and ribonucelic acid and deoxyribonucleic acid content of tissues. J. Lab. Clin. Med. 77:144-152, 1971.

1285a. Prasad, A. S., D. Oberleas, and C. Rajasekeran. Essential micronutrient elements. Biochemistry and changes in liver disorders. Amer. J. Clin. Nutr. 23:581-591, 1970.

1286. Prasad, A. S., D. Oberleas, P. Wolf, and J. P. Horwitz. Studies on zinc deficiency: Changes in trace elements and enzyme activities in tissues of zinc-deficient rats. J. Clin. Invest. 46:549-557, 1967.

1287. Prasad, A. S., H. H. Sandstead, A. R. Schulert, and A. S. El Rooby. Urinary cretion of zinc in patients with the syndrome of anemia, hepatosplenomegaly, dwarfism, and hypogonadism. J. Lab. Clin. Med. 62:591-599, 1963.

1287a. Prasad, A. S., E. B. Schoomaker, J. Ortega, G. J. Brewer, D. Oberleas, and F. J. Oelshlegel, Jr. Zinc deficiency in sickle cell disease. Clin. Chem. 21:582-587, 1975.

1288. Prasad, A. S., A. R. Schulert, H. H. Sandstead, A. Miale, Jr., and Z. Farid. Zinc, iron and nitrogen content of sweat in normal and deficient subjects. J. Lab. Clin. Med. 62:84-89, 1963.

1289. Prask, J. A., and D. J. Plocke. A role for zinc in structural integrity of cytoplasmic ribosomes of Euglena gracilis. Plant Physiol. 48: 150-155, 1971.

1290. Preston, A. The concentration of ^{65}Zn in the flesh of oysters related to the discharge of cooling pond effluent from the C.E.G.B. Nuclear Power Station at Bradwell-on-Sea, Essex, pp. 995-1004. In B. Åberg and F. P. Hungate, Eds. Radioecological Concentration Processes. Proceedings of an International Symposium held in Stockholm 25-29 April, 1966. New York: Pergamon, 1967.

1291. Preston, A., D. F. Jefferies, J. W. R. Dutton, B. R. Harvey, and A. K. Steele. British Isles coastal waters: The concentrations of selected heavy metals in sea water, suspended matter, and biological indicators--a pilot survey. Environ. Pollut. 3:69-82, 1972.

1292. Pretlow, T. P., and F. Sherman. Porphyrins and zinc porphyrins in normal and mutant strains of yeast. Biochim. Biophys. Acta 148:629-644, 1967.

1293. Price, C. A. A zinc-dependent lactate dehydrogenase in Euglena gracilis. Biochem. J. 82:61-66, 1962.

1294. Price, C. A. Control of processes sensitive to zinc in plants and microorganisms, pp. 69-89. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1295. Price, C. A. Molecular Approaches to Plant Physiology. New York: McGraw-Hill, 1970. 398 pp.

1296. Price, C. A., H. E. Clark, and E. A. Funkhouser. Functions of micronutrients in plants, pp. 231-242. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

1297. Price, C. A., and E. Millar. Zinc, growth, & respiration in euglena. Plant Physiol. 37:423-427, 1962.

1298. Price, N. O., and G. E. Bunce. Effect of nitrogen and calcium on balance of copper, manganese, and zinc in preadolescent girls. Nutr. Rep. Int. 5:275-280, 1972.

1299. Price, N. O., G. E. Bunce, and R. W. Engel. Copper, manganese, and zinc balance in preadolescent girls. Amer. J. Clin. Nutr. 23:258-260, 1970.

1300. Price, W. J. Analytical Atomic Absorption Spectrometry. New York: Heyden and Son, Ltd., 1972. 239 pp.

1301. Pringle, B. H. D. E. Hissong, E. L. Katz, and S. T. Mulawka. Trace metal accumulation by estuarine mollusks. J. Sanit. Eng. Div. Proc. Amer. Soc. Civil Eng. 94(SA3):455-475, 1968.

1302. Pringle, W. L., W. K. Dawley, and J. E. Miltimore. Sufficiency of Cu and Zn in barley, forage, and corn silage rations as measured by response to supplements by beef cattle. Can. J. Anim. Sci. 53:497-502, 1973.

1303. Prohaska, J. R., R. W. Luecke, and R. Jasinski. Effect of zinc deficiency from day 18 of gestation and/or during lactation on the development of some rat brain enzymes. J. Nutr. 104:1525-1531, 1974.

1304. Pulido, P., J. H. R. Kägi, and B. L. Vallee. Isolation and some properties of human metallothionein. Biochemistry 5:1768-1777, 1966.

1305. Pullen, F. W., II. Oral zinc and vitamin therapy for laryngotracheal trauma and surgical aftercare, pp. 237-242. In W. J. Pories, J. M. Hsu, and R. L. Woosley, Eds. Clinical Applications of Zinc Metabolism. Proceedings of the International Symposium. Springfield, Ill.: Charles C Thomas, 1974.

1305a. Pullen, F. W. Postintubation tracheal granuloma. Arch. Otolaryngol. 92: 340-342, 1970.

1305b. Pullen, F. W., II., J. Pories, and W. H. Strain. Delayed healing: The rationale for zinc therapy. Laryngoscope 81:1638-1649, 1971.

1306. Pulliam, H. R., G. W. Barrett, and E. P. Odum. Bioelimination of tracer ^{65}Zn in relation to metabolic rates in mice, pp. 725-730. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn: U. S. Atomic Energy Commission, 1969.

1306a. Purichia, N., and L. C. Erway. Effects of dichlorophenamide, zinc, and manganese on otolith development in mice. Dev. Biol. 27:395-405, 1972.

1306b. Pyatayev, G. Ye. State of the olfactory function in zinc production workers. Zh. Ushn. Nos. Gorl. Bolezn. 31(4):17-21, 1971. (in Russian, summary in English)

1307. Quarantillo, E. P., Jr. Effect of supplemental zinc on wound healing in rats. Amer. J. Surg. 121:661-664, 1971.

1307a. Quarterman, J. The effects of zinc deficiency or excess on the adrenals and the thymus in the rat, pp. 742-744. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium on Trace Element Metabolism in Animals, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

1307b. Quarterman, J. The effect of zinc on uptake of glucose by adipose tissue. Biochim. Biophys. Acta 177:644-646, 1969.

1307c. Quarterman, J. Trace element deficiencies: Variation among species, pp. 17-23. In W. D. Tavernor, Ed. Nutrition and Disease in Experimental Animals. Proceedings of a Symposium, London, May 1969. London: Baillière, Tindall & Cassell, 1970.

1308. Quarterman, J., and E. Florence. Observations on glucose tolerance and plasma levels of free fatty acid and insulin in zinc-deficient rat. Brit. J. Nutr. 28:75-79, 1972.

1309. Quarterman, J., W. R. Humphries, J. Morrison, and F. A. Jackson. The effect of zinc deficiency on intestinal and salivary mucins. Biochem. Soc. Trans. 1:101, 1973.

1309a. Quarterman, J., C. F. Mills, and W. R. Humphries. The reduced secretion of, and sensitivity to insulin in zinc-deficient rats. Biochem. Biophys. Res. Commun. 25:354-358, 1966.

1310. Rachlin, L. Aortic zinc in patients with peripheral vascular disease. Angiology 23:651-655, 1972.

1311. Rahn, K. A., R. Dams, J. A. Robbins, and J. W. Winchester. Diurnal variations of aerosol trace element concentrations as determined by nondestructive neutron activation analysis. Atmos. Environ. 5:413-422, 1971.

1311a. Rand, M. C., A. E. Greenberg, and M. J. Taras, Eds. Zinc, pp. 144-146, 148-151, and 262-269. In Standard Methods for the Examination of Water and Wastewater. (14th ed.) Washington, D. C.: American Public Health Association, 1976.

1312. Randall, P. J., and D. Bouma. Zinc deficiency, carbonic anhydrase, and photosynthesis in leaves of spinach. Plant Physiol. 52:229-232, 1973.

1313. Randoin, L., P. Le Gallic, Y. Dupuis, and A. Barnardin. Tables de Composition des Aliments. Paris: Jacques Lanore, 1961. 116 pp.

1314. Rankhawa, N. S., and F. E. Broadbent. Soil organic matter-metal complexes: 5. Reactions of zinc with model compounds and humic acid. Soil Sci. 99:295-300, 1965.

1315. Randhawa, N. S., and F. E. Broadbent. Soil organic matter-metal complexes: 6. Stability constants of zinc-humic acid complexes at different pH values. Soil Sci. 99:362-366, 1965.

1316. Ranweiler, L. E., and J. L. Moyers. Atomic absorption procedure for analysis of metals in atmospheric particulate matter. Environ. Sci. Technol. 8:152-156, 1974.

1317. Rathore, V. S., Y. P. S. Bajaj, and S. H. Wittwer. Subcellular localization of zinc and calcium in bean (Phaseolus vulgaris L.) tissues. Plant Physiol. 49:207-211, 1972.

1318. Rathore, V. S., S. H. Wittwer, W. H. Jyung, Y. P. S. Bajaj, and M. W. Adams. Mechanisms of zinc uptake in bean (Phaseolus vulgaris) tissue. Physiol. Plant. 23:908-919, 1970.

1319. Raulin, J. Études chémiques sur la végetation. Ann. Sci. Nat. (5th Series) Bot. 11:93-299, 1869.

1320. Rauterberg, E., and W. Bussler. Anreicherung von Zink im Boden und Schäden durch Zink an Planzen in einem Drahthaus. Z. Pflanzenernähr. Düng. Bodenk. 106:35-38, 1964.

1321. Reed, H. S. Effects of zinc deficiency on phosphate metabolism of the tomato plant. Amer. J. Bot. 33:778-784, 1946.

1322. Reid, T. W., and I. B. Wilson. E. coli alkaline phosphatase, pp. 373-415. In P. D. Boyer, Ed. The Enzymes. Vol. 4. Hydrolysis. Other C-N Bonds Phosphate Esters. (3rd ed.) New York: Academic Press, 1971.

1323. Reimann, E. M., M. L. Sunde, and W. G. Hoekstra. Effect of chloroquine and certain amines, vitamins, and arthritis-influencing agents on the zinc-deficient chick. Proc. Soc. Exp. Biol. Med. 137:473-479, 1971.

1324. Reimann, E. M., M. L. Sunde, and W. G. Hoekstra. Effect of zinc, certain related dietary factors and age on the histamine content of chick tissues. J. Nutr. 101:1623-1630, 1971.

1325. Reinhold, J. G. High phytate content of rural Iranian bread: A possible cause of human zinc deficiency. Amer. J. Clin. Nutr. 24:1204-1206, 1971.

1326. Reinhold, J. G. Phytate destruction by yeast fermentation in whole wheat meals. J. Amer. Diet. Assoc. 66:38-41, 1975.

1326a. Reinhold, J. G., and G. A. Kfoury. Zinc-dependent enzymes in zinc-depleted rats; intestinal alkaline phosphatase. Amer. J. Clin. Nutr. 22:1250-1263, 1969.

1327. Reinhold, J. G., G. A. Kfoury, and T. A. Thomas. Zinc, copper and iron concentrations in hair and other tissues: Effects of low zinc and low protein intakes in rats. J. Nutr. 92:173-182, 1967.

1328. Reinhold, J. G., K. Nasr, A. Lahimgarzadeh, and H. Hedayati. Effects of purified phytate and phytate-rich bread upon metabolism of zinc, calcium, phosphorus, and nitrogen in man. Lancet 1:283-288, 1973.

1329. Reinhold, J. G., A. Parsa, N. Karimian, J. W. Hammick, and F. Ismail-Beigi. Availability of zinc in leavened and unleavened wholemeal wheaten breads as measured by solubility and uptake by rat intestine in vitro. J. Nutr. 104:976-982, 1974.

1330. Reinhold, J. G., E. Pascoe, M. Arslanian, and K. Bitar. Relation of zinc metalloenzyme activities to zinc concentrations in tissues. Biochim. Biophys. Acta 215:430-437, 1970.

1331. Reinhold, J. G., and H. A. Ronaghy. Zinc deficiency in man. Lancet 2:50, 1973. (letter)

1332. Renfro, W. C. Radioecology of Zinc-65 in an Arm of the Columbia River Estuary. Ph.D. Thesis. Corvallis: Oregon State University, 1968. 94 pp.

1332a. Renfro, W. C., and C. Osterberg. Radiozinc decline in starry flounders after temporary shut down of Hanford reactors, pp. 372-379. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Symposium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

1333. Delete 1333--use 28a.

1334. Reshetkina, L. P. On the content of zinc, iron, copper and cobalt in some products entering the childrens' diet in Prikarpatie. Vopr. Pitan. 24(5):68-72, 1965. (in Russian, summary in English)

1335. Reusch, C. S., and L. D. Bunch. Serum zinc level in diabetes. J.A.M.A. 210:2285, 1969. (letter)

1336. Revúsová, V., V. Zvara, and J. Gratzlová. Urinary zinc excretion in patients with urolithiasis. Urol. Int. 28:72-79, 1973.

1337. Rhodes, J. R., A. H. Pradzynski, C. B. Hunter, J. S. Payne, and J. L. Lindgren. Energy dispersive x-ray fluorescence analysis of air particulates in Texas. Environ. Sci. Technol. 6:922-927, 1972.

1338. Rice, E. W. Morphological changes in human spermatozoa following treatment of semen with certain dialkyldithiocarbamates. Exp. Cell Res. 34:186-188, 1964.

1339. Rice, T. R. Review of zinc in ecology, pp. 619-631. In V. Schultz, and A. W. Klement, Jr., Eds. Radioecology. New York: Reinhold, 1963.

1340. Riceman, D. S., and G. B. Jones. Distribution of zinc and copper in seedlings of subterranean clover (Trifolium subterraneum L.) in solution culture. Austral. J. Agric. Res. 7:495-503, 1956.

1341. Riceman, D. S., and G. B. Jones. Distribution of zinc in subterraneum clover (Trifolium subterraneum L.) grown to maturity in a culture solution containing zinc labelled with the radioactive isotope ^{65}Zn. Austral. J. Agric. Res. 9:730-744, 1958.

1342. Richards, M. P., and R. J. Cousins. Influence of parenteral zinc and actinomycin D on tissue zinc uptake and the synthesis of a zinc-binding protein. Bioinorgan. Chem. 4:215-224, 1975.

1343. Richards, M. P., and R. J. Cousins. Zinc binding protein: Relationship to hepatic metabolism. Fed. Proc. 34:906, 1975. (abstract)

1344. Richards, N. J., and D. G. Williams. Complex formation between aqueous zinc chloride and cellulose-related D-glucopyranosides. Carbohydr. Res. 12:409-420, 1970.

1345. Richards, O. C., and W. J. Rutter. Preparation and properties of yeast aldolase. J. Biol. Chem. 236:3177-3185, 1961.

1345a. Rickard, B. R. Facial eczema: Zinc responsiveness in dairy cattle. N. Z. Vet. J. 23:41-42, 1975.

1345b. Rickwood, D., and H. G. Klemperer. Decreased ribonucleic acid synthesis in isolated rat liver nuclei during starvation. Biochem. J. 120: 381-384, 1970.

1345c. Richmond, C. R., J. E. Furchner, G. A. Trafton, and W. H. Langham. Comparative metabolism of radionuclides in mammals--I. Uptake and retention of orally administered Zn^{65} by four mammalian species. Health Phys. 8:481-489, 1962.

1345d. Delete--use 1345c.

1346. Riepe, M. E., and J. H. Wang. Elucidation of the catalytic mechanism of carbonic anhydrase. J. Amer. Chem. Soc. 89:4229, 1967. (letter)

1347. Riepe, M. E., and J. H. Wang. Infrared studies on the mechanism of action of carbonic anhydrase. J. Biol. Chem. 243:2779-2787, 1968.

1348. Riesner, K., and G. Seifert. Häufigkeit, Lokalisation und Pathogenese der Herzgerüstverkalkung. Virchows Arch. (Path. Anat) 355:1-18, 1972.

1349. Riley, J. P., and D. Taylor. The concentrations of cadmium, copper, iron, manganese, molybdenum, nickel, vanadium and zinc in part of the tropical north-east Atlantic Ocean. Deep-Sea Res. 19:307-317, 1972.

1349a. Rimington, C. Patterns of porphyrin excretion and their interpretation. S. Afr. J. Lab. Clin. Med. 9:255-261, 1963.

1350. Rivière, M. R., I. Chouroulinkov, and M. Guérin. Production de tumerus par injections intratesticulaires de chlorure de zinc chez le rat. Bull. Assoc. Fr. Etude Cancer 47:55-87, 1960.

1351. Roberson, R. H., and P. J. Schaible. The tolerance of growing chicks for high levels of different forms of zinc. Poult. Sci. 39:893-896, 1960.

1352. Roberts, K. R., W. J. Miller, P. E. Stake, R. P. Gentry, and M. W. Neathery. High dietary cadmium on zinc absorption and metabolism in calves fed for comparable nitrogen balances. Proc. Soc. Exp. Biol. Med. 144: 906-908, 1973.

1353. Robertson, D. E. Contamination problems in trace-element analysis and ultrapurification, pp. 207-253. In M. Zief and R. Speights, Eds. Ultrapurity: Methods and Techniques. New York: Marcel Dekker, Inc., 1972.

1354. Robinson, M. F., J. M. McKenzie, C. D. Thomson, and A. L. van Rij. Metabolic balance of zinc, copper, cadmium, iron, molybdenum and selenium in young New Zealand women. Brit. J. Nutr. 30:195-205, 1973.

1354a. Zinc Institute, Inc. Annual Review 1974. U. S. Zinc Industry Including Statements from Other Countries. New York: Zinc Institute, Inc., 1975. 29 pp.

1355. Robinson, S., and A. H. Robinson. Chemical composition of sweat. Physiol. Rev. 34:202-220, 1954.

1355a. Rodin, A. E., and A. S. Goldman. Autopsy findings in acrodermatitis enteropathica. Amer. J. Clin. Path. 51:315-322, 1969.

1356. Roels, O. A., and N. S. T. Lui. Vitamin A and carotene, pp. 142-157. In R. S. Goodhart and M. E. Shils, Eds. Modern Nutrition in Health and Disease. Dietotherapy. (5th ed.) Philadelphia: Lea & Febiger, 1973.

1357. Rogers, G. R. Collaborative study of the atomic absorption spectrophotometric method for determining zinc in foods. J. Assoc. Off. Anal. Chem. 51:1042-1045, 1968.

1358. Rohrs, L. C. Metal fume fever from inhaling zinc oxide. A.M.A. Arch. Ind. Health 16:42-47, 1957.

1359. Rolfe, G. L., J. W. Melin, and B. B. Ewing. Lead pollution in a watershed ecosystem. Paper Presented at American Institute of Biological Sciences Symposium on Impact of Heavy Metals on Ecosystems, Minneapolis, Minnesoto, 1972.

1360. Roman, W. Zinc in porphyria. Amer. J. Clin. Nutr. 22:1290-1303, 1969.

1361. Romeril, M. G. The uptake and distribution of ^{65}Zn in oysters. Marine Biol. 9:347-354, 1971.

1362. Ronaghy, H., R. M. S. Fox, S. M. Garn, H. Israel, A. Harp, P. G. Moe, and J. A. Halsted. Controlled zinc supplementation for malnourished school boys: A pilot experiment. Amer. J. Clin. Nutr. 22:1279-1289, 1969.

1363. Ronaghy, H. A., J. G. Reinhold, M. Mahloudji, P. Ghavami, M. R. S. Fox, and J. A. Halsted. Zinc supplementation of malnourished schoolboys in Iran: Increased growth and other effects. Amer. J. Clin. Nutr. 27: 112-121, 1974.

1363a. Roschin, A. V., and L. A. Timofeevskaya. Chemical substances in the work environment: Some comparative aspects of USSR and US hygienic standards. Ambio 4:30-33, 1975.

1364. Rose, F. L., and C. E. Cushing. Periphyton: Autoradiography of zinc-65 adsorption. Science 168:576-577, 1970.

1365. Rosenblum, D., and S. J. Petzold. Granulocyte alkaline phosphatase. Studies of purified enzymes from normal subjects and patients with polycythemia vera. J. Clin. Invest. 52:1665-1672, 1973.

1366. Rosman, K. J. R. A survey of the isotopic and elemental abundance of zinc. Geochim. Cosmochim. Acta 36:801-819, 1972.

1367. Rosoff, B., and C. R. Martin. Effect of gonadotrophins and of testosterone on organ weights and zinc-65 uptake in the male rat. Gen. Comp. Endocrinol. 10:75-84, 1968.

1368. Rosoff, B., and H. Spencer. Tissue distribution of zinc-65 in tumour tissue and normal tissue in man. Nature 207:652-653, 1965.

1369. Rosner, F., and P. C. Gorfien. Erythrocyte and plasma zinc and magnesium levels in health and disease. J. Lab. Clin. Med. 72:213-219, 1968.

1370. Roth, H.-P., and M. Kirchgessner. Aktivitätsveränderungen verschiedener Dehydrogenasen und der alkalischen Phosphatase im Serum bei Zink-Depletion und -Repletion. 6. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 32:289-296, 1974.

1371. Roth, H.-P., and M. Kirchgessner. De- und Repletionsstudien an Zink in Knocken und Leber wachsender Ratten. Arch. Tierernaehr. 24:283-298, 1974.

1372. Roth, H.-P., and M. Kirchgessner. Zum Aktivitätsverlauf verschiedener Dehydrogenasen in der Rattenleber bei unterschiedlicher Zinkversorgung. 8. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 33:1-9, 1974.

1373. Roth, H.-P., and M. Kirchgessner. Zum Einfluss unterschiedlicher Diätzinkgehalte auf die Aktivität der alkalischen Phosphatase im Knochen. 9. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 33:57-61, 1974.

1374. Roth, H.-P., and M. Kirchgessner. Zur Aktivität der Blut-Carboanhydrase bei Zinkmangel wachsender Ratten. 7. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 32:296-300, 1974.

1375. Roth, H.-P., and M. Kirchgessner. Zur Aktivität der Pankreas-carboxypeptidase A und B bei Zink-Depletion und -Repletion. 10. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 33:62-66, 1974.

1376. Roth, H.-P., and M. Kirchgessner. Zur Enzymaktivität von Dehydrogenasen im Rattenmuskel bei Zinkmangel. 11. Zum Stoffwechsel des Zinks im tierischen Organismus. Z. Tierphysiol. 33:67-71, 1974.

1376a. Rowett Research Institute. Annual Report of Studies in Animal Nutrition and Allied Sciences. Vol. 27. Bucksburn, Aberdeen: Rowett Research Institute, United Kingdom Agricultural Research Council, 1971.

1376b. Roy, S. K., B. S. Setty, H. Chandra, and A. B. Kar. The effect of efferent duct ligation on the uptake of ^{65}Zn by the epididymis andy vas deferens of Rhesus monkeys (Macaca mulatta). Acta Endocrinol. 77:186-192, 1974.

1377. Rubin, H. Inhibition of DNA synthesis in animal cells by ethylene diamine tetraacetate, and its reversal by zinc. Proc. Nat. Acad. Sci. U.S.A. 69:712-716, 1972.

1378. Rubin, H. pH, serum and Zn^{++} in the regulation of DNA synthesis in cultures of chick embryo cells. J. Cell. Physiol. 82:231-238, 1973.

1379. Rubin, H., and T. Koide. Inhibition of DNA synthesis in chick embryo cultures by deprivation of either serum or zinc. J. Cell Biol. 56:777-786, 1973.

1380. Rubin, H., and T. Koide. Stimulation of DNA synthesis and 2-deoxy-d-glucose transport in chick embryo cultures by excessive metal concentrations and by a carcinogenic hydrocarbon. J. Cell. Physiol. 81: 387-396, 1973.

1381. Rubini, M. E., G. Montalvo, C. P. Lockhart, and C. R. Johnson. Metabolism of zinc-65. Amer. J. Physiol. 200:1345-1348, 1961.

1382. Rubins, E. J. Soils, pp. 1865-1880. In F. J. Welcher, Ed. Standard Methods of Chemical Analysis. Vol. 3. Instrumental Methods. Part B. (6th ed.) New York: D. Van Nostrand Company, Inc., 1966.

1382a. Ruch, R. R., H. J. Gluskoter, and N. F. Shimp. Occurrence and Distribution of Potentially Volatile Trace Elements in Coal: A Final Report. Environmental Geology Notes 72. Urbana: Illinois State Geology Survey, 1974. 96 pp.

1383. Rucker, J. B., and J. W. Valentine. Salinity response of trace element concentration in Crassostrea virginica. Nature 190:1099-1100, 1961.

1384. Rudgers, L. A., J. L. Demeterio, G. M. Paulsen, and R. Ellis, Jr. Interaction among atrazine, temperature, and phosphorus-induced zinc deficiency in corn (Zea mays, L.). Soil Sci. Soc. Amer. Proc. 34:240-248, 1970.

1385. Rule, A. H., L. Kopito, and H. Shwachman. Chemical analyses of ejaculates from patients with cystic fibrosis. Fertil. Steril. 21:515-520, 1970.

1386. Rush, V. A., and V. V. Lizunova. The chemical composition of the cedar nut. Vopr. Pitan. 26(3):93, 1967. (in Russian)

1387. Ryan, P., J. Lee, and T. F. Peebles. Trace Element Problems in Relation to Soil Units in Europe. (Working Party on Soil Classification and Survey of the European Commission on Agriculture). World Soil Resources Report No. 31. Rome: Food and Agriculture Organization of the United Nations, 1967. 55 pp.

1388. Sabath, L. D., and M. Finland. Thiol-group binding of zinc to a β-lactamase of Bacillus cereus: Differential effects on enzyme activity with penicillin and cephalosporins as substrates. J. Bacteriol. 95:1513-1519, 1968.

1389. Sabodash, V. M. Dynamics of zinc concentration in the early developmental stages of carp. Hydrobiol. J. 6(3):59-65, 1970.

1390. Sacks, R. D., and S. W. Brewer, Jr. Metals analysis in particulate pollutants by emission spectroscophy. Appl. Spectrosc. Rev. 6:313-349, 1972.

1391. Sadasivan, V. Studies on the biochemistry of zinc. 1. Effect of feeding zinc on the liver and bones of rats. Biochem. J. 48:527-530, 1951.

1392. Saito, S., L. Zeitz, I. M. Bush, R. Lee, and W. F. Whitmore, Jr. Zinc content of spermatozoa from various levels of canine and rat reproductive tracts. Amer. J. Physiol. 213:749-752, 1967.

1393. Saito, S., L. Zeitz, I. M. Bush, R. Lee, and W. F. Whitmore, Jr. Zinc uptake in canine or rat spermatozoa. Amer. J. Physiol. 217:1038-1043, 1969.

1394. Salami, A. U., and D. G. Kenefick. Stimulation of growth in zinc-deficient corn seedlings by the addition of tryptophan. Crop Sci. 10:291-294, 1970.

1395. Saldeen, T., and U. Brunk. Enzyme histochemical investigations of the inhibitory effect of zinc on the injurious action of carbon tetrachloride on the liver. Frankfurter Z. Path. 76:419-426, 1967.

1396. Saldeen, T., and U. Stenram. Uptake of Zn^{65} by rat liver damaged by CCl_4. Frankfurter Z. Path. 77:61-66, 1967.

1397. Salo, E. O., and W. L. Leet. The concentration of ^{65}Zn by oysters maintained in the discharge canal of a nuclear power plant, pp. 363-371. In D. J. Nelson and F. C. Evans, Eds. Symposium on Radioecology. Proceedings of the Second National Sympsoium held at Ann Arbor, Michigan, May 15-17, 1967. CONF-670503. TID-4500. Oak Ridge, Tenn.: U. S. Atomic Energy Commission, 1969.

1398. Saltman, P., and H. Boroughs. The accumulation of zinc by fish liver slices. Arch. Biochem. Biophys. 86:159-174, 1960.

1399. Samachson, J., J. Dennis, R. Fowler, and A. Schmitz. The reaction of ^{65}Zn with surfaces of bone and bone mineral. Biochim. Biophys. Acta 148:767-773, 1967.

1399a. Samachson, J., and A. Schmitz. The reactions of H^+ and Zn^{2+} with the surfaces of bone and bone mineral. Biochim. Biophys. Acta 170: 409-419, 1968.

1400. Sampson, J., R. Graham, and H. R. Hester. Studies on feeding zinc to pigs. Cornell Vet. 32:225-236, 1942.

1401. Sandell, E. B. Determination of copper, zinc and lead in silicate rocks. Ind. Eng. Chem. Anal. Ed. 9:464-469, 1937.

1402. Sandell, E. B. [Dithizone analysis], pp. 144-176, and Zinc, pp. 941-965. In Colorimetric Determination of Traces of Metals. (3rd ed.) New York: Interscience Publishers, Inc., 1959.

1402a. Sandow, A., and A. Isaacson. Effects of methylene blue, acridine orange, and zinc on muscular contraction. Biochem. Biophys. Res. Commun. 2:455-458, 1960.

1402b. Sandow, A., and S. M. Bien. Blockade of neuromuscular transmission by zinc. Nature 193:689-690, 1962.

1403. Delete--use 1408.

1403a. Sandstead, H. H. Effect of chronic lead intoxication on <u>in vivo</u> I^{131} uptake by the rat thyroid. Proc. Soc. Exp. Biol. Med. 124:18-20, 1967.

1403b. Sandstead, H. H. Cadmium, zinc, and lead, pp. 43-56. In Geochemistry and the Environment. Vol. 1. The Relation of Selected Trace Elements to Health and Disease. Washington, D. C.: National Academy of Sciences, 1974.

1404. Sandstead, H. H. Zinc nutrition in the United States. Amer. J. Clin. Nutr. 26:1251-1260, 1973.

1404a. Sandstead, H. H., R. F. Burk, G. H. Booth, Jr., and W. J. Darby. Current concepts on trace minerals. Clinical considerations. Med. Clin. North Amer. 54:1509-1531, 1970.

1405. Sandstead, H. H., Y. Y. Al-Ubaidi, E. Halas, and G. Fosmire. Zinc deficiency during the critical period for brain growth, pp. 745-748. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium, 1973. Baltimore: University Park Press, 1974.

1406. Sandstead, H. H., G. J. Fosmire, J. M. McKenzie, and E. S. Halas. Zinc deficiency and brain development in the rat. Fed. Proc. 34:86-88, 1975.

1407. Sandstead, H. H., D. D. Gillespie, and R. N. Brady. Zinc deficiency: Effect on brain of the suckling rat. Pediatr. Res. 6:119-125, 1972.

1408. Sandstead, H. H., V. C. Lanier, Jr., G. H. Shephard, and D. D. Gillespie. Zinc and wound healing. Effects of zinc deficiency and zinc supplementation. Amer. J. Clin. Nutr. 23:514-519, 1970.

1409. Sandstead, H. H., A. S. Prasad, Z. Farid, A. Schulert, A. Miale, Jr., S. Bassilly, and W. J. Darby. Endocrine manifestations of human zinc deficiency, pp. 304-320. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1410. Sandstead, H. H., A. S. Prasad, A. R. Schulert, Z. Farid, A. Miale, Jr., S. Bassilly, and W. J. Darby. Human zinc deficiency, endocrine manifestations and response to treatment. Amer. J. Clin. Nutr. 20: 422-442, 1967.

1410a. Sandstead, H. H., and R. A. Rinaldi. Impairment of deoxyribonucleic acid synthesis by dietary zinc deficiency in the rat. J. Cell Physiol. 73:81-83, 1969.

1411. Sandstead, H. H., and G. H. Shepard. The effect of zinc deficiency on the tensile strength of healing surgical incisions in the integument of the rat. Proc. Soc. Exp. Biol. Med. 128:687-689, 1968.

1412. Sandstead, H. H., A. S. Shukry, A. S. Prasad, M. K. Gabr, A. El Hifney, N. Mokhtar, and W. J. Darby. Kwashiorkor in Egypt. I. Clinical and biochemical studies, with special reference to plasma zinc and serum lactic dehydrogenase. Amer. J. Clin. Nutr. 17:15-26, 1965.

1412a. Sanyal, S. N. An experimental study on the treatment of enlarged prostate. J. Indian Med. Assoc. 63:276-278, 1974.

1413. Saraswat, R. D., and S. P. Arora. Effect of dietary zinc on the vitamin A level and alkaline phosphatase activity in blood sera of lambs. Indian J. Anim. Sci. 42:358-362, 1972.

1414. Sarram, M., M. Younessi, P. Knorvath, G. A. Kfoury, and J. G. Reinhold. Zinc nutrition in human pregnancy in Fars Province, Iran. Significance of geographic and socioeconomic factors. Amer. J. Clin. Nutr. 22:726-732, 1969.

1414a. Sartor, J. D., and G. B. Boyd. Water Pollution Aspects of Street Surface Contaminants. EPA-R2-72-081. Washington, D. C.: U. S. Government Printing Office, 1972. 236 pp.

1415. Sato, N., and R. I. Henkin. Pituitary-gonadal regulation of copper and zinc metabolism in the female rat. Amer. J. Physiol. 225:508-512, 1973.

1416. Sauchelli, V. Copper, pp. 151-171. In Trace Elements in Agriculture. New York: Van Nostrand Reinhold Co., 1969.

1417. Sauchelli, V. Iron, pp. 39-57. In Trace Elements in Agriculture. New York: Van Nostrand Reinhold Co., 1969.

1418. Savlov, E. D., W. H. Strain, and F. Huegin. Radiozinc studies in experimental wound healing. J. Surg. Res. 2:209-212, 1962.

1419. Sayre, J. D. Accumulation of Radioisotopes in Corn Leaves. Research Bull. 723. Wooster: Ohio Agricultural Experiment Station, 1952. 30 pp.

1420. Schall, H., and H. Schall, Jr. Nahrungsmitteltabelle-Tabelle. (18th ed.) Leipzig: Johann Ambrosius Barth, 1962. 196 pp.

1420a. Schechter, P. J., E. L. Giroux, J. L. Schlienger, V. Hoenig, and A. Sjoerdsma. Distribution of serum zinc between albumin and α_2-macroglobulin in patients with decompensated hepatic cirrhosis. Eur. J. Clin. Invest. 6:147-150, 1976.

1420b. Schechter, P. J., W. T. Friedewald, D. A. Bronzert, M. S. Raff, and R. I. Henkin. Idopathic hypogeusia: A description of the syndrome and a single-blind study with zinc sulfate. Int. Rev. Neurobiol. Suppl. 1: 125-140, 1972.

1421. Schelling, J. L., S. Muller-Hess, and F. Thonney. Effect of food on zinc absorption. Lancet 2:969-969, 1973.

1422. Schelske, C. L. Fallout ^{54}Mn accumulated by bay scallops Argopecten irradians (Lamarck) near Beaufort, North Carolina, pp. 331-346. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA in Seattle, U.S.A., 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

1423. Schicha, H., K. Kasperek, H. J. Klein, and L. F. Feinendegen. Bestimmung von Spurenelementkonzentrationen in Hirnrinde und Basalganglien beim Menschen und ihre Abhängigkeit vom Lebensalter. Fortschr. Gebiet Röntgenstrahl. Nuklearmed. 1972(Suppl.):184-185, 1972.

1424. Schicha, H., W. Müller, K. Kasperek, and R. Schröder. Neutronenaktivierungsanalytische Bestimmung des Spurenelementgehaltes in operativ entnommenem Gewebe des menschlichen Grosshirnes. Beitr. Path. 146:366-374, 1972.

1425. Schlettwein-Gsell, D., and S. Mommsen-Straub. Übersicht Spurenelemente in Lebensmitteln. I. Zink. Int. Z. Vit. Forsch. 40:659-572, 1970.

1426. Schlicker, S. A., and D. H. Cox. Maternal dietary zinc, and development and zinc, iron, and copper content of the rat fetus. J. Nutr. 95: 287-294, 1968.

1427. Schmid, K., and W. Bürgi. Preparation and properties of the human plasma Ba-α_2-glycoproteins. Biochim. Biophys. Acta 47:440-453, 1961.

1428. Schmid, W. E., H. P. Haag, and E. Epstein. Absorption of zinc by excised barley roots. Physiol. Plant. 18:860-869, 1965.

1429. Schmid, K., and S. Takahashi. Polymorphism of zinc-α_2-human glycoprotein. Nature 203:407-408, 1964.

1430. Schmidt-Kehl, L. Wie kann Zinkoxyd bei einatmung Fieber erzeugen? (Mechanismus der Giessfieberentstehung) Zentr. Gewerbehyg. Unfallverhüt. 5:272-273, 1928.

1431. Schmitt, N., G. Brown, E. L. Devlin, A. A. Larsen, E. D. McCausland, and J. M. Saville. Lead poisoning in horses. An environmental health hazard. Arch. Environ. Health 23:185-195, 1971.

1432. Schnappinger, M. G., Jr., D. C. Martens, G. W. Hawkins, D. F. Amos, and G. D. McCart. Response of corn to residual and applied zinc as $ZnSO_4$ and Zn-EDTA in field investigations. Agron. J. 64:64-66, 1972.

1433. Schneider, E., and C. A. Price. Decreased ribonucleic acid levels: A possible cause of growth inhibition in zinc deficiency. Biochim. Biophys. Acta 55:406-408, 1962.

1434. Schneider, R. F. The Impact of Various Heavy Metals on the Aquatic Environment. Technical Report Number 2. Denver: U. S. Environmental Protection Agency. Water Quality Office, 1971. 26 pp.

1435. Schnitzer, M., and S. U. Khan. Humic Substances in the Environment. New York: Marcel Dekker, Inc., 1972. 327 pp.

1436. Schnitzer, M., and S. I. M. Skinner. Organo-metallic interactions in soils: 5. Stability constants of Cu^{++}-, Fe^{++}-, and Zn^{++}- fulvic acid complexes. Soil Sci. 102:361-365, 1966.

1437. Schor, R. A., S. G. Prussin, D. L. Jewett, J. J. Ludowieg, and R. S. Bhatnagar. Trace levels of manganese, copper, and zinc in rib cartilage or related to age in humans and animals, both normal and dwarfed. Clin. Orthopaed. Related Res. 93:346-355, 1973.

1438. Schormüller, J., Ed. Hanbuch der Lebensmittelchemie. Vol. 6. Alkaloidhaltige. Genussmittel, Gewürze, Kochsalz. Berlin: Springer-Verlag, 1967. 770 pp.

1439. Schraer, K. K., and D. H. Calloway. Zinc balance in pregnant teenagers. Nutr. Metabol. 17:205-212, 1974.

1440. Schramel, P., K. Samsahl, and J. Pavlu. Determination of 12 selected microelements in air particles by neutron activation analysis. J. Radioanal. Chem. 19:329-337, 1974.

1440a. Schreiner, G. E., J. F. Maher, R. B. Freeman, and J. M. B. O'Connell. Problems of hemodialysis, pp. 316-332. In E. L. Becker, Ed. Proceedings of 3rd International Congress of Nephrology, Washington, D. C., 1966. Vol. 3. Chemical Nephrology. New York: S. Karger, 1967.

1441. Schrenk, H. H., H. Heimann, G. D. Clayton, W. M. Gafafer, and H. Wexler. Air Pollution in Donora, Pa. Epidemiology of the Unusual Smog Episode of October 1948. Preliminary Report. Public Health Bulletin No. 306. Washington, D. C.: Federal Security Agency, Public Health Service, 1949. 173 pp.

1442. Delete 1442--same as 1542

1443. Schrodt, G. R., T. Hall, and W. F. Whitmore, Jr. The concentration of zinc in diseased human prostate glands. Cancer 17:1555-1566, 1964.

1444. Schroeder, H. A. Cadmium as a factor in hypertension. J. Chron. Dis. 18:647-656, 1965.

1445. Schroeder, H. A. Cadmium, chromium, and cardiovascular disease. Circulation 35:570-582, 1967.

1446. Schroeder, H. A. Losses of vitamins and trace minerals resulting from processing and preservation of foods. Amer. J. Clin. Nutr. 24: 562-573, 1971.

1446a. Schroeder, H. A. Relation between mortality from cardiovascular disease and treated water supplies. J.A.M.A. 172:1902-1908, 1960.

1447. Schroeder, H. A. The role of chromium in mammalian nutrition. Amer. J. Clin. Nutr. 21:230-244, 1968.

1447a. Schroeder, H. A., J. J. Balassa, and I. H. Tipton. Essential trace metals in man: Manganese. A study in homeostasis. J. Chron. Dis. 19:545-571, 1966.

1448. Schroeder, H. A., and J. Buckman. Cadmium hypertension. Its reversal in rats by a zinc chelate. Arch. Environ. Health 14:693-697, 1967.

1449. Schroeder, H. A., and M. Mitchener. Toxic effects of trace elements on the reproduction of mice and rats. Arch. Environ. Health 23:102-106, 1971.

1450. Schroeder, H. A., and A. P. Nason. Trace metals in human hair. J. Invest. Derm. 53:71-78, 1969.

1451. Schroeder, H. A., A. P. Nason, I. H. Tipton, and J. J. Balassa. Essential trace metals in man: Zinc. Relation to environmental cadmium. J. Chron. Dis. 20:179-210, 1967.

1451a. Schryver, H. F., H. F. Hintz, J. E. Lowe, R. L. Hintz, R. B. Harper, and J. T. Reid. Mineral composition of the whole body, liver and bone of young horses. J. Nutr. 104:126-132, 1974.

1451b. Schuhmann, R., Jr., and H. W. Schadler. Chemistry and physics of zinc technology. pp. 65-102. In C. H. Mathewson, Ed. Zinc. The Science and Technology of the Metal, its Alloys and Compounds. New York: Reinhold Publishing Corporation, 1959.

1452. Schuster, I., and E. Broda. Die Bindung von Zink durch Zellwände von Chlorella. Monatsh. Chem. 101:285-295, 1970.

1453. Schütte, K. H. The Biology of Trace Elements. Their Role in Nutrition. Philadelphia: J. B. Lippincott Co., 1964. 228 pp.

1454. Schwarz, F. J., and M. Kirchgessner. Absorption von Zink-65 und Kupfer-64 im Zinkmangel. Int. J. Vit. Nutr. Res. 44:258-266, 1974.

1455. Schwarz, F. J., and M. Kirchgessner. In-vitro-Untersuchungen zur intestinalen Zink-Absorption. Z. Tierphysiol. 34:67-76, 1974.

1456. Schwarz, F. J., and M. Kirchgessner. Wechselwirkungen bei der intestinalen Absorption von ^{64}Cu, ^{65}Zn und ^{59}Fe nach Cu-, Zn- oder Fe-Depletion. Int. Z. Vit. Ernährungsforsch. 44:117-126, 1973.

1457. Schweigart, H. A. Vitalstoff-Lehre. Vitalstoff-Tabellarium. Dachau-Munich: Verlag Hans Zauner, Jr., 1962. 230 pp.

1457a. Scott, D. A. Crystalline insulin. Biochem. J. 28:1592-1602, 1934.

1458. Scott, D. A., and A. M. Fisher. Studies on the pancreas and liver of normal and of zinc-fed cats. Amer. J. Physiol. 121:253-260, 1938.

1459. Scott, D. A., and A. M. Fisher. The effect of zinc salts on the action of insulin. J. Pharmacol. Exp. Ther. 55:206-221, 1935.

1460. Scott, D. A., and A. M. Fisher. The insulin and the zinc content of normal and diabetic pancreas. J. Clin. Invest. 17:725-728, 1938.

1461. Scott, D. B. M., and M. E. Sano. Disruption of the estrous cycle of the hamster on a zinc-deficient diet. Fed. Proc. 34:940, 1975. (abstract)

1462. Scrutton, M. C., C. W. Wu, and D. A. Goldthwait. Presence and possible role of zinc in RNA polymerase obtained from Escherichia coli. Proc. Nat. Acad. Sci. U.S.A. 68:2497-2501, 1971.

1463. Scrutton, M. C., M. R. Young, and M. F. Utter. Pyruvate carboxylase from bakers' yeast. J. Biol. Chem. 245:6220-6227, 1970.

1464. Seatz, L. F., and J. J. Jurinak. Zinc and soil fertility, pp. 115-121. In A. Stefferud, Ed. Soil. U. S. Department of Agriculture. The Yearbook of Agriculture 1957. Washington, D. C.: U. S. Government Printing Office, 1957.

1465. Sedberry, J. E., Jr., N. B. Lieu, F. J. Peterson, and F. E. Wilson. The effects of applications of zinc and different sources of phosphorus on growth and nutrient uptake of rice. Commun. Soil Sci. Plant Anal. 4: 259-267, 1973.

1466. Seeley, J. L., and R. K. Skogerboe. Combined sampling-analysis method for the determination of trace elements in atmospheric particulates. Anal. Chem. 46:415-421, 1974.

1466a. Sefton, G., R. G. Clark, and G. Owen. Changes in serum zinc after operation. Brit. J. Surg. 61:329, 1974. (abstract)

1467. Segar, D. A., J. D. Collins, and J. P. Riley. The distribution of the major and some minor elements in marine animals. Part II. Molluscs. J. Marine Biol. Assoc. U. K. 51:131-136, 1971.

1468. Seidel, A. Comparison of the effectiveness of CaDTPA and ZnDTPA in removing ^{241}Am from the rat. Radiat. Res. 54:304-315, 1973.

1469. Seidel, A., and V. Volf. Removal of internally deposited transuranium elements by Zn-DTPA. Health Phys. 22:779-783, 1972.

1470. Seifter, S., S. Takahashi, and E. Harper. Further demonstration that cysteine reacts with the metal component of collagenase. Biochim. Biophys. Acta 214:559-561, 1970.

1471. Sereda, G. A., and V. A. Artemova. Determination of zinc and lead in the atmosphere by means of preliminary chromatographic fractioning. Gig. Sanit. 38(8):62-64, 1973. (in Russian)

1471a. Serjeant, G. R., R. E. Galloway, and M. C. Gueri. Oral zinc sulphate in sickle-cell ulcers. Lancet 2:891-892, 1970.

1472. Settlemire, C. T., and G. Matrone. In vivo effect of zinc on iron turnover in rats and life span of the erythrocyte. J. Nutr. 92:159-164, 1967.

1473. Settlemire, C. T., and G. Matrone. In vivo interference of zinc with ferritin iron in the rat. J. Nutr. 92:153-158, 1967.

1474. Seutter, E., and A. H. M. Sutorius. The quantitative analysis of some constituents of crude sweat. Dermatologica 145:203-209, 1972.

1474a. Sever, L. E. Zinc and human development: A review. Human Ecol. 3: 43-57, 1975.

1475. Sever, L. E., and I. Emanuel. Is there a connection between maternal zinc deficiency and congenital malformations of the central nervous system in man? Teratology 7:117-118, 1973. (letter)

1476. Seymour, A. H. Accumulation and loss of zinc-65 by oysters in a natural environment, pp. 605-619. In Disposal of Radioactive Wastes into Seas, Oceans and Surface Waters. Proceedings of a Symposium, Vienna 16-20 May 1966. Vienna: International Atomic Energy Agency, 1966.

1477. Seymour, A. H., and V. A. Nelson. Decline of ^{65}Zn in marine mussels following the shutdown of Hanford reactors, pp. 277-286. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA in Seattle, U.S.A., 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

1477a. Shacklette, H. T., J. C. Hamilton, J. G. Boerngen, and J. M. Bowles. Elemental Composition of Surficial Materials in the Conterminous United States. U. S. Geological Survey Professional Paper 574-D.. Washington, D. C.: U. S. Government Printing Office, 1971. 71 pp.

1478. Shah, B. G., J. C. Meranger, G. Belonje, and D. S. Deshmukh. Zinc retention in young rats fed increasing levels of high-zinc oysters. J. Fish. Res. Board Can. 28:843-848, 1971.

1479. Shanklin, S. H., E. R. Miller, D. E. Ullrey, J. A. Hoefer, and R. W. Luecke. Zinc requirement of baby pigs on casein diets. J. Nutr. 96:101-108, 1968.

1480. Shaw, N. A., H. C. Dickey, H. H. Brugman, D. L. Blamberg, and J. F. Witter. Zinc deficiency in female rabbits. Lab. Anim. 8:1-7, 1974.

1481. Sheline, G. E., I. L. Chaikoff, H. B. Jones, and M. L. Montgomery. Studies on the metabolism of zinc with the aid of its radioactive isotope. I. The excretion of administered zinc in urine and feces. J. Biol. Chem. 147:404-414, 1943.

1481a. Sherlock, S. Diseases of the Liver and Biliary System. (4th ed.) Philadelphia: F. A. Davis Company, 1968. 809 pp.

1482. Shevchuk, I. A. Variations of zinc content and distribution in the pancreas following alloxan and insulin administration. Fed. Proc. 24(Translation Suppl.): 48-50, 1965.

1482a. Shevchuk, I. A. Zinc metabolism and the activity of alkaline phosphatase in the kidneys of albino rats in alloxan diabetes and in repeated administration of insulin. Prob. Endokrinol. (Mosk) 19(1):61-65, 1973. (in Russian, summary in English)

1483. Shevchuk, I. A., and L. I. Sandulyak. The content and functional histotopography of zinc in the kidneys. Byull. Eksp. Biol. Med. 73(12): 44-47, 1972. (in Russian, summary in English)

1483a. Shils, M. E., W. L. Wright, A. Turnbull, and F. Brescia. Long-term parenteral nutrition through an external arteriovenous shunt. New Engl. J. Med. 283:341-344, 1970.

1484. Shin, Y. A., and G. L. Eichhorn. Interactions of metal ions with polynucleotides and related compounds. XI. The reversible unwinding and rewinding of deoxyribonucleic acid by zinc (II) ions through temperature manipulation. Biochemistry 7:1026-1032, 1968.

1485. Shrader, R. E., and L. S. Hurley. Enzyme histochemistry of peripheral blood and bone marrow in zinc-deficient rats. Lab. Invest. 26:566-571, 1972.

1486. Shulman, J., I. L. Brisbin, and W. Knox. Effect of temperature, salinity, and food intake on the excretion of Zn^{65} in small marine fish. Biol. Bull. 121:378, 1961. (abstract)

1487. Shuster, C. N., Jr., and B. H. Pringle. Trace metal accumulation by the American oyster, Crassostrea virginica. Proc. Nat. Shellfish. Assoc. 59:91-103, 1969.

1488. Sibly, P. M., and J. G. Wood. The nature of carbonic anhydrase from plant sources. Austral. J. Sci. Res. B 4:500-510, 1951.

1488a. Siegel, E., F. A. Graig, M. M. Crystal, and E. P. Siegel. Distribution of ^{65}Zn in the prostate and other organs of man. Brit. J. Cancer 15:647-664, 1961.

1489. Sinha, E. Metals as Pollutants in Air and Water. An Annotated Bibliography. Ocean Engineering Information Series Vol. 6. La Jolla, Calif.: Ocean Engineering Information Service, 1972. 91 pp.

1490. Skidmore, J. F. Toxicity of zinc compounds to aquatic animals, with special reference to fish. Q. Rev. Biol. 39:227-248, 1964.

1491. Skoog, F. Relationships between zinc and auxin in the growth of higher plants. Amer. J. Bot. 27:939-951, 1940.

1491a. Short, E. M., L. J. Elsas, and L. E. Rosenberg. Effect of parathyroid hormone on renal tubular reabsorption of amino acids. Metabolism 23:715-727, 1974.

1492. Shukla, U. C., and H. Raj. Influence of genetic variability on zinc response in wheat. Soil Sci. Soc. Amer. Proc. 38:477-479, 1974.

1493. Skulachev, V. P., V. V. Chistyakov, A. A. Jasaitis, and E. G. Smirnova. Inhibition of the respiratory chain by zinc ions. Biochem. Biophys. Res. Commun. 26:1-6, 1967.

1494. Slater, J. P., A. S. Mildvan, and L. A. Loeb. Zinc in DNA polymerases. Biochem. Biophys. Res. Commun. 44:37-43, 1971.

1495. Slavik, M., D. Danilson, H. R. Keiser, and R. I. Henkin. Alterations in metabolism of copper and zinc after administration of 6-azauridine triacetate. Biochem. Pharm. 22:2349-2352, 1973.

1496. Slavik, M., W. Lovenberg, and H. R. Keiser. Changes in serum and urine amino acids in patients with progressive systemic sclerosis treated with 6-azauridine triacetate. Biochem. Pharm. 22:1295-1300, 1973.

1497. Slobodien, M. J., A. Brodsky, C. H. Ke, and I. Horm. Removal of zinc from humans by DTPA chelation therapy. Health Phys. 24:327-330, 1973.

1498. Slowey, J. F., and D. W. Hood. Copper, manganese and zinc concentrations in Gulf of Mexico waters. Geochim. Cosmochim. Acta 35:121-138, 1971.

1499. Small, L. F. Experimental studies on the transfer of ^{65}Zn in high concentration by euphausiids. J. Exp. Marine Biol. Ecol. 3:106-123, 1969.

1500. Small, L. F., and S. W. Fowler. Turnover and vertical transport of zinc by the euphausiid Meganyctiphanes norvegica in the Ligurian Sea. Marine Biol. 18:284-290, 1973.

1501. Small, L. F., S. W. Fowler, and S. Keckes. Flux of zinc through a macroplanktonic crustacean, pp. 437-452. In Radioactive Contamination of the Marine Environment. Proceedings, Symposium on Radioactive Contamination of Marine Environment, Seattle, U. S. A., 1972. Vienna: International Atomic Energy Agency, 1973.

1502. Smilde, K. W., P. Konkoulakis, and B. van Luit. Crop response to phosphate and lime on acid sandy soils high in zinc. Plant Soil 41:445-457, 1974.

1503. Smit, Z. M. Studies in the metabolism of zinc. III. The urinary excretion of zinc in adult white and Bantu subjects. S. Afr. J. Lab. Clin. Med. 8:59-62, 1962.

1504. Smit, Z. M., and P. J. Pretorius. Studies in metabolism of zinc. Part 2. Serum zinc levels and urinary zinc excretions in South African Bantu Kwashiorkor patients. J. Trop. Pediatr. 9:105-112, 1964.

1505. Smith, F. R., R. I. Henkin, and R. B. Dell. Disordered gustatory acuity in liver disease. Gastroenterology 70:568-571, 1976.

1506. Smith, A. G., and L. Powell. Genesis of teratomas of the testis. A study of the normal and zinc-injected testes of roosters. Amer. J. Path. 33:653-670, 1957.

1507. Smith, D. L., and W. G. Schrenk. Application of atomic absorption spectroscopy to plant analysis. I. Comparison of zinc and manganese analysis with official AOAC colorimetric methods. J. Assoc. Off. Anal. Chem. 55:669-675, 1972.

1508. Smith, I. D., R. H. Grummer, W. G. Hoekstra, and P. H. Phillips. Effect of feeding an autoclaved diet on the development of parakeratosis in swine. J. Anim. Sci. 19:568-579, 1960.

1509. Smith, J. C., E. G. Daniel, L. D. McBean, F. S. Doft, and J. A. Halsted. Effect of microorganisms on zinc metabolism, p. 353. In P. L. White, Ed. Proceedings. Western Hemisphere Nutrition Congress III. August 30 - September 2, 1971, Miami Beach, Florida. Mount Kisco, N. Y.: Futura Publishing Company, Inc., 1972. (abstract)

1510. Smith, J. C., Jr., and J. A. Halsted. Clay ingestion (geophagia) as a source of zinc for rats. J. Nutr. 100:973-980, 1970.

1511. Smith, J. C., Jr., E. G. McDaniel, W. Chan, and E. D. Brown. Effect of zinc therapy and factors associated with growth on vitamin A metabolism. Fed. Proc. 34:907, 1975. (abstract)

1512. Smith, J. C., Jr., E. G. McDaniel, F. F. Fan, and J. A. Halsted. Zinc: A trace element essential in vitamin A metabolism. Science 181: 954-955, 1973.

1513. Smith, J. C., Jr., E. G. McDaniel, L. D. McBean, F. S. Doft, and J. A. Halsted. Effect of microorganisms upon zinc metabolism using germfree and conventional rats. J. Nutr. 102:711-720, 1972.

1514. Smith, J. E., E. D. Brown, and J. C. Smith, Jr. The effect of zinc deficiency on the metabolism of retinol-binding protein in the rat. J. Lab. Clin. Med. 84:692-697, 1974.

1515. Smith, J. M. Zinc, Alkaline Phosphatase, and Heat-Stable Alkaline Phosphatase in the Plasma of "High Risk" Pregnant Women in East Harlem, New York: A Longitudinal Study. M. S. Thesis. Ithaca, N. Y.: Cornell University, 1972.

1516. Smith, S. E., and E. J. Larson. Zinc toxicity in rats. Antagonistic effects of copper and liver. J. Biol. Chem. 163:29-38, 1946.

1517. Smith, W. H., M. P. Plumlee, and W. M. Beeson. Effect of source of protein on zinc requirement of the growing pig. J. Anim. Sci. 21: 399-405, 1962.

1517a. Snaith, S. M., A. J. Hay, and G. A. Levvy. The relationship between the α-mannosidase activity and the zinc content of mammalian sex organs. J. Endocrinol. 50:659-667, 1971.

1517b. Smyth, C. J., A. G. Lasichak, and S. Levey. The effect of orally and intravenously administered amino acid mixtures on voluntary food consumption in normal men. J. Clin. Invest. 26:439-445, 1947.

1518. Snaith, S. M., and G. A. Levvy. Purification and properties of α-D-mannosidase from Jack-Bean meal. Biochem. J. 110:663-670, 1968.

1519. Snaith, S. M., and G. A. Levvy. Purification and properties of α-D-mannosidase from rat epididymis. Biochem. J. 114:25-33, 1969.

1520. Snaith, S. M., G. A. Levvy, and A. J. Hay. Purification and properties of α-D-mannosidase from the limpet Patella vulgata. Biochem. J. 117: 129-137, 1970.

1520a. Sneddon, I. Acrodermatitis enteropathica. Brit. J. Derm. 91:716, 1974. (letter)

1521. Snell, F. D., and C. T. Snell. Lead [dithizone methods], pp. 1-7, and Zinc by dithizone, pp. 412-419. In Colorimetric Methods of Analysis Vol. 2. Inorganic. (3rd ed.) Princeton, N. J.: D. Van Nostrand Company, Inc., 1949.

1521a. Solomons, N. W., I. H. Rosenberg, and H. H. Sandstead. Zinc nutrition in celiac sprue. Amer. J. Clin. Nutr. 29:371-375, 1976.

1521b. Solomons, N. W., T. J. Layden, I. H. Rosenberg, K. Vo-Khactu, and H. H. Sandstead. Plasma trace metals during total parenteral alimentation. Gastroenterology 70:1022-1025, 1976.

1522. Solomons, N. W., K. V. Khactu, H. H. Sandstead, and I. H. Rosenberg. Zinc nutrition in regional enteritis (RE). Clin. Res. 22:582A, 1974. (abstract)

1522a. Somers, M., and E. J. Underwood. Ribonuclease activity and nucleic acid and protein metabolism in the testes of zinc-deficient rats. Austral. J. Biol. Sci. 22:1277-1282, 1969.

1522b. Soman, S. D., K. T. Joseph, S. J. Raut, C. D. Mulay, M. Parameshwaran, and V. K. Panday. Studies on major and trace element content in human tissues. Health Phys. 19:641-656, 1970.

1523. Sommer, A. L., and C. B. Lipman. Evidence on the indispensible nature of zinc for higher green plants. Plant Physiol. 1:231-249, 1926.

1524. Sonneland, J. The "inoperable" breast carcinoma. A successful result using zinc chloride fixative. Amer. J. Surg. 124:391-393, 1972.

1525. Sorenson, J. R. J., E. G. Melby, P. J. Nord, and H. G. Petering. Interferences in the determination of metallic elements in human hair. An evaluation of zinc, copper, lead, and cadmium, using atomic absorption spectrophotometry. Arch. Environ. Health 27:36-39, 1973.

1526. Souci, S. W., W. Fachmann, and H. Kraut. Die Zusammensetzung der Lebensmittel. Nährwert-Tabellen. Stuttgart: Wissenschaftliche Verlagsgesellschaft M. B. H., 1962.

1527. Spadaro, J. A., R. O. Becker, and C. H. Bachman. The distribution of trace metal ions in bone and tendon. Calcif. Tissue Res. 6:49-54, 1970.

1528. Spais, A. G., and A. A. Papasteriadis. Zinc deficiency in cattle under Greek conditions, pp. 628-631. In W. G. Hoekstra, J. W. Suttie, H. E. Ganther, and W. Mertz, Eds. Trace Element Metabolism in Animals - 2. Proceedings of the Second International Symposium on Trace Element Metabolism in Animals, held in Madison, Wisconsin, 1973. Baltimore: University Park Press, 1974.

1529. Sparks, R. E., W. T. Waller, and J. Cairns, Jr. Effect of shelters on the resistance of dominant and submissive bluegills (Lepomis macrochirus) to a lethal concentration of zinc. J. Fish. Res. Board Can. 29:1356-1358, 1972.

1530. Spencer, D. W., and P. G. Brewer. Distribution of copper, zinc and nickel in sea water of the Gulf of Maine and the Sargasso Sea. Geochim. Cosmochim. Acta 33:325-339, 1969.

1531. Spencer, D. W., P. G. Brewer, and P. L. Sachs. Aspects of the distribution and trace element composition of suspended matter in the Black Sea. Geochim. Cosmochim. Acta 36:71-86, 1972.

1532. Spencer, H., D. Osis, L. Kramer, and C. Norris. Studies of zinc metabolism in man, pp. 193-204. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - V. Proceedings of 5th Annual Conference on Trace Substances in Environmental Health held in Columbia, Missouri, June 29 - July 1, 1971. Columbia: University of Missouri, 1972.

1533. Spencer, H., and B. Rosoff. Effect of chelating agents on the removal of zinc-65 in man. Health Phys. 12:475-480, 1966.

1534. Spencer, H., B. Rosoff, A. Feldstein, S. H. Cohn, and E. Gusmano. Metabolism of zinc-65 in man. Radiat. Res. 24:432-445, 1965.

1535. Spencer, H., B. Rosoff, I. Lewin, and J. Samachson. Studies of zinc-65 metabolism in man, pp. 339-362. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1536. Spencer, H., and J. Samachson. Secretion of alkaline earth metals in saliva, pp. 101-113. In L. H. Schneyer and C. A. Schneyer, Eds. Secretory Mechanisms of Salivary Glands. Proceedings of an International Conference on Mechanisms of Salivary Secretion and Their Regulation held at the University of Alabama Medical Center, Birmingham, Alabama, August 9-11, 1966. New York: Academic Press, 1967.

1537. Spencer, H., and J. Samachson. Studies of zinc metabolism in man, pp. 312-314. In C. F. Mills, Ed. Trace Element Metabolism in Animals. Proceedings of WAAP/IBP International Symposium, Aberdeen, Scotland, July 1969. London: E. & S. Livingston, 1970.

1538. Spencer, H., V. Vankinscott, I. Lewin, and J. Samachson. Zinc-65 metabolism during low and high calcium intake in man. J. Nutr. 86:169-177, 1965.

1539. Sporn, A., I. Dinu, L. Stoenescu, and A. Cirstea. Beiträge zur Ermittlung der Wechselwirkungen zwischen Cadmium und Zink. Nahrung 13:461-469, 1969.

1540. Sprague, J. B. Promising anti-pollutant: Chelating agent NTA protects fish from copper and zinc. Nature 220:1345-1346, 1968.

1541. Sprague, J. B., and B. A. Ramsay. Lethal levels of mixed copper-zinc solutions for juvenile salmon. J. Fish. Res. Board Can. 22:425-432, 1965.

1542. Springgate, C. F., A. S. Mildvan, R. Abramson, J. L. Engle, and L. A. Loeb. Escherichia coli deoxyribonucleic acid polymerase I, a zinc metalloenzyme. J. Biol. Chem. 248:5987-5993, 1973.

1543. Stake, P. E., W. J. Miller, and R. P. Gentry. Zinc metabolism and homeostasis in ruminants as affected by dietary energy intake and growth rate. Proc. Soc. Exp. Biol. Med. 142:494-496, 1973.

1544. Stake, P. E., W. J. Miller, R. P. Gentry, and M. W. Neathery. Zinc metabolic adaptations in calves fed a high but nontoxic zinc level for varying time periods. J. Anim. Sci. 40:132-137, 1975.

1545. Stake, P. E., W. J. Miller, M. W. Neathery, and R. P. Gentry. Zinc-65 absorption and tissue distribution in two- and six-month-old Holstein calves and lactating cows. J. Dairy Sci. 58:78-81, 1975.

1546. Staker, E. V., and R. W. Cumming. The influence of zinc on the productivity of certain New York peat soils. Soil Sci. Soc. Amer. Proc. 6:207-214, 1941.

1547. Starcher, B. C. Studies on the mechanism of copper absorption in the chick. J. Nutr. 97:321-326, 1969.

1547a. Steele, T. H. Dissociation of zinc excretion from other cations in man. J. Lab. Clin. Med. 81:205-213, 1973.

1548. Stefanini, U. Sul contenuto in zinco del siero materno e fetale e della placenta nella gravidanza normale a termine. Ann. Ostet. Ginecol. 86:771-775, 1964.

1549. Stein, W. D. Chemical composition of the melanin granule and its relation to the mitochondrion. Nature 175:256-257, 1955.

1550. Stephan, J. K., and J. M. Hsu. Effect of zinc deficiency and wounding on DNA synthesis in rat skin. J. Nutr. 103:548-552, 1973.

1551. Stern, A., M. Nalder, and I. G. Macy. Zinc retention in childhood. J. Nutr. 21(Suppl.):8, 1941. (abstract)

1552. Stern, K., and R. Willheim. [Effect of zinc on cancerous tissue], p. 40. In The Biochemistry of Malignant Tumors. Brooklyn, N.Y.: Chemical Publishing Co., Inc., 1943.

1553. Stevenson, F. J., and M. S. Ardakani. Organic matter reactions involving micronutrients in soils, pp. 79-114. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsey, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

1554. Stewart, I., C. D. Leonard, and G. Edwards. Factors influencing the absorption of zinc by citrus. Florida State Hort. Soc. Proc. 68: 82-88, 1955.

1554a. Stöber, M. Parakeratose beim schwarzbunten Niederungskalb. 1. Klinisches Bild und Ätiologie. Dtsch. Tieraerztl. Wochenschr. 78:257-265, 1971.

1555. Stokinger, H. E. /Zinc oxide fume/, p. 1185. In D. W. Fassett and D. D. Irish, Eds. Toxicology. Vol. 2. In F. A. Patty, Ed. Industrial Hygiene and Toxicology (2nd ed.) New York: Interscience Publishers, 1963.

1555a. Strain, W. H., A. Lascari, and W. J. Pories. Zinc deficiency in babies, pp. 759-765. In Proceedings of the Seventh International Congress on Nutrition. Vol. 5. Physiology and Biochemistry of Food Components. New York: Pergamon Press, 1967.

1556. Strain, W. H., E. G. Mansour, A. Flynne, W. J. Pories, A. J. Tomaro, and O. A. Hill, Jr. Plasma-zinc concentration in patients with bronchogenic cancer. Lancet 1:1021-1022, 1972. (letter)

1557. Strain, W. H., and W. J. Pories. Zinc levels of hair as tools in zinc metabolism, pp. 363-377. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1557a. Strain, W. H., C. G. Rob, W. J. Pories, R. C. Childers, M. F. Thompson, Jr., J. A. Hennessen, and F. M. Grater. Element imbalances of atherosclertic aortas, pp. 128-133. In J. R. Devoe, Ed. Modern Trends in Activation Analysis. Proceedings of the 1968 International Conference held at the National Bureau of Standards, Gaithersburg, Maryland, Oct. 7-11, 1968. NBS Special Publication 312. Vol. I. Washington, D. C.: U. S. Government Printing Office, 1969.

1558. Strain, W. H., L. T. Steadman, C. A. Lankau, W. P. Berliner, and W. J. Pories. Analysis of zinc levels in hair for the diagnosis of zinc deficiency in man. J. Lab. Clin. Med. 68:244-249, 1966.

1558a. Strauss, J. S., and P. E. Pochi. The quantitative gravimetric determination of sebum production. J. Invest. Derm. 36:293-298, 1961.

1559. Stukenholtz, D. C., R. J. Olsen, G. Gogan, and R. A. Olson. On the mechanism of phosphorus-zinc interaction in corn nutrition. Soil Sci. Soc. Amer. Proc. 30:759-763, 1966.

1560. Sudakova, N. M. Effect of the trace element zinc on some indexes of lipid metabolism in experimental animals. Uch. Zap. Petrozavodsk. Gos. Univ. 19:333-336, 1972. (in Russian)

1561. Sudia, T. W., and D. G. Green. The translocation of Zn^{65} and Cs^{134} between seed generations in soybean (Glycine max (L.) Merr.) Plant Soil 37: 695-697, 1972.

1562. Sugimae, A. Emission spectrographic determination of trace elements in airborne particulate matter collected on silver membrane filter. Appl. Spectrosc. 28:458-461, 1974.

1563. Sulima, S. Ya. The blood content of iron, copper, cobalt and zinc in ulcerous stenosis of the pylorus. Klin. Med. 43(2):62-66, 1965. (in Russian, summary in English)

1564. Sullivan, J. F. Effect of alcohol on urinary zinc excretion. Q. J. Stud. Alcohol 23:216-220, 1962.

1565. Sullivan, J. F., R. E. Burch, H. J. Quigley, and D. F. Magee. Zinc deficiency and decreased pancreatic secretory response. Amer. J. Physiol. 227:105-108, 1974.

1566. Sullivan, J. F., and R. P. Heaney. Zinc metabolism in alcoholic liver disease. Amer. J. Clin. Nutr. 23:170-177, 1970.

1567. Sullivan, J. F., and R. P. Heaney. Zinc-65 metabolism in patients with cirrhosis. J. Lab. Clin. Med. 66:1025, 1965. (abstract)

1568. Sullivan, J. F., and H. G. Lankford. Zinc metabolism and chronic alcoholism. Amer. J. Clin. Nutr. 17:57-63, 1965.

1569. Sullivan, J. F., J. O'Grady, and H. G. Lankford. The zinc content of pancreatic secretion. Gastroenterology 48:438-443, 1965.

1570. Summerell, J. M., A. Osmand, and G. H. Smith. An equilibrium-dialysis study of the binding of zinc to insulin. Biochem. J. 95:31P, 1965. (abstract)

1571. Sumner, J. B., and K. Myrbäck, Eds. The Enzymes. Vol. 1, Part 1. Chemistry and Mechanism of Action. New York: Academic Press, 1950. 724 pp.

1572. Sunde, M. L. Zinc requirement for normal feathering of commercial leghorn-type pullets. Poult. Sci. 51:1316-1322, 1972.

1572a. Sunderman, F. W., Jr. Current status of zinc deficiency in the pathogenesis of neurological, dermatological and musculoskeletal disorders. Ann. Clin. Lab. Sci. 5:132-145, 1975.

1573. Sunderman, F. W., Jr. Metal carcinogenesis in experimental animals. Food Cosmet. Toxicol. 9:105-120, 1971.

1574. Supplee, W. C. Antagonistic relationship between dietary cadmium and zinc. Science 139:119-120, 1963.

1575. Supplee, W. C. Production of zinc deficiency in turkey poults by dietary cadmium. Poult. Sci. 40:827-828, 1961.

1576. Suso, F. A., and H. M. Edwards, Jr. Binding capacity of intestinal mucosa and blood plasma for zinc. Proc. Soc. Exp. Biol. Med. 137: 306-309, 1971.

1577. Suso, F. A., and H. M. Edwards, Jr. Ethylenediaminetetraacetic acid and ^{65}Zn binding by intestinal digesta, intestinal mucosa and blood plasma. Proc. Soc. Exp. Biol. Med. 138:157-162, 1971.

1578. Sutton, W. R., and V. E. Nelson. Blood sugar changes in the rat produced by salts of beryllium, magnesium, and zinc with some observations on hemoglobin and red blood corpuscles. Proc. Iowa Acad. Sci. 45:115-121, 1938.

1579. Sutton, W. R., and V. E. Nelson. Growth, reproduction and blood changes produced in rats by means of zinc carbonate. Proc. Iowa Acad. Sci. 44:117-121, 1937.

1580. Sutton, W. R., and V. E. Nelson. Studies on zinc. Proc. Soc. Exp. Biol. Med. 36:211-213, 1937.

1581. Swaine, D. J. The Trace-Element Content of Soils. Commonwealth Bureau of Soil Science Technical Communication No. 48. Farnham Royal Bucks, England: Commonwealth Agricultural Bureaux, 1955. 157 pp.

1582. Swenerton, H., and L. S. Hurley. Teratogenic effects of a chelating agent and their prevention by zinc. Science 173:62-63, 1971.

1583. Swenerton, H., R. Shrader, and L. S. Hurley. Lactic and malic dehydrogenases in testes of zinc-deficient rats. Proc. Soc. Exp. Biol. Med. 141:283-286, 1972.

1584. Swenerton, H., R. Shrader, and L. S. Hurley. Zinc-deficient embryos: Reduced thymidine incorporation. Science 166:1014-1015, 1969.

1585. Swiller, A. I., and H. E. Swiller. Metal fume fever. Amer. J. Med. 22: 173-174, 1957.

1585a. Szadkowski, D., H.-P. Ehrhardt, K.-H. Schaller, and G. Lehnert. Der Zink-und Kupferspiegel im Serum hitzeadaptierter Schwerarbeiter. Int. Arch. Gewerbepath. Gewerbehyg. 25:202-214, 1969.

1586. Szmigielski, S. Hypothetical significance of disturbances of zinc and protoporphyrin metabolism in leukaemic cells. Nature 209:411-412, 1966.

1587. Szmigielski, S., and J. Litwin. Histochemical demonstration of zinc in blood granulocytes — a new test in diagnosis of neoplastic diseases. Cancer 17:1381-1384, 1964.

1588. Szmigielski, S., and J. Litwin. The histochemical study of zinc content in granulocytes in normal adults and in hematologic disorders. Blood 25:56-62, 1965.

1589. Szmigielski, S., and J. Litwin. Zinc content in polymorphonuclear leukocytes of healthy subjects and patients with certain blood diseases. Pol. Arch. Med. Wewn. 34:319-321, 1964. (in Polish, summary in English)

1590. Tabor, E. C., and J. E. Meeker. Effects of the 1956 Steel Strike on Air Pollution Levels in Several Communities. Paper 58-24 Presented at the 51st Annual Meeting of the Air Pollution Control Association, May 25-29, 1958. 20 pp.

1591. Takino, M. Calcium, magnesium, zinc and iron in fishes' meat. Bol. Ind. Anim. (São Paulo) 24:285-292, 1967. (in Portuguese)

1592. Talbot, T. R., Jr., and J. F. Ross. Zinc content of plasma and erythrocytes of patients with pernicious anemia, sickle cell anemia, polycythemia vera, leukemia and neoplastic disease. Lab. Invest. 9: 174-184, 1960.

1593. Tallamy, P. T., and H. E. Randolph. Influence of mastitis on properties of milk. V. Total and free concentration of major minerals in skimmilk. J. Dairy Sci. 53:1386-1388, 1970.

1594. Tallmadge, J. A. Non-ferrous metals, pp. 255-283. In C. F. Gurnham, Ed. Industrial Wastewater Control. New York: Academic Press, 1965.

1594a. Tamasi, G. C., and E. J. Drenick. Resistance to insulin convulsions in fasted mice. Endocrinology 92:1277-1279, 1973.

1595. Tan, Y. T. Composition and nutritive value of some grasses, plants and aquatic weeds tested as diets. J. Fish Biol. 2:253-257, 1970.

1596. Tanis, R. J., R. E. Tashian, and Y.-S. L. Yu. Properties of carbonic anhydrase isozymes isolated from porcine erythrocytes. J. Biol. Chem. 245:6003-6009, 1970.

1597. Tanner, J. T., J. P. F. Lambert, and R. E. Simpson. The neutron activation analysis program of the Food and Drug Administration. J. Assoc. Off. Anal. Chem. 53:1140-1144, 1970.

1598. Tao, S.-H., and L. S. Hurley. Changes in plasma proteins in zinc-deficient rats. Proc. Soc. Exp. Biol. Med. 136:165-167, 1971.

1599. Tao, S.-H., and L. S. Hurley. Effect of dietary calcium deficiency during pregnancy on zinc mobilization in intact and parathyroidectomized rats. J. Nutr. 105:220-225, 1975.

1600. Deleted.

1601. Deleted.

1601a. Tatum, H. J. Metallic copper as an intrauterine contraceptive agent. Amer. J. Obstet. Gynecol. 117:602-618, 1973.

1602. Tauber, F. W., and A. C. Krause. The role of iron, copper, zinc, and manganese in the metabolism of the ocular tissues, with special reference to the lens. Amer. J. Ophthalmol. 26:260-266, 1943.

1603. Terhune, M. W., and H. H. Sandstead. Decreased RNA polymerase activity in mammalian zinc deficiency. Science 177:68-69, 1972.

1604. Terman, G. L., P. M. Giordano, and S. E. Allen. Relationships between dry matter yields and concentrations of Zn and P in young corn plants. Agron. J. 64:684-687, 1972.

1605. Terry, C. W., B. E. Terry, and J. Davies. Transfer of zinc^{65} across the placenta and fetal membranes of the rabbit. Amer. J. Physiol. 198: 303-308, 1960.

1606. Thiers, R. E. Contamination in trace element analysis and its control. Methods Biochem. Anal. 5:273-335, 1957.

1607. Theorell, H., A. P. Nygaard, and R. Bonnichsen. Studies on liver alcohol dehydrogenase. III. The influence of pH and some anions on the reaction velocity constants. Acta Chem. Scand. 9:1148-1165, 1955.

1607a. Theuer, R. C., and W. G. Hoekstra. Oxidation of ^{14}C-labeled carbohydrate, fat, and amino acid substrates by zinc-deficient rats. J. Nutr. 89: 448-454, 1966.

1607b. Thomas, H. R., S. J. Lahoda, J. D. Hedges, and E. T. Kornegay. Zinc requirement for gestating-lactating sows. Virginia Polytech. Inst. Res. Div. Rep. 158:156-158, 1974.

1607c. Thompson, A. P. Industrial zinc oxide, zinc sulfide and other zinc compounds, pp. 637-668. In C. H. Mathewson, Ed. Zinc: The Science and Technology of the Metal, Its Alloys and Compounds. New York: Reinhold Publishing Corporation, 1959.

1608. Thompson, R. C. 7(h) Elmešu, elmušu, SÙ.UD.ÁG.(GÁ), brass, pp. 76-79. In A Dictionary of Assyrian Chemistry and Geology. Oxford: The Clarendon Press, 1936.

1609. Thompson, R. J., G. B. Morgan, and L. J. Purdue. Analysis of selected elements in atmospheric particulate matter by atomic absorption, pp. 178-188. In J. W. Scales, Ed. Air Quality Instrumentation. Vol. 1. Pittsburgh: Instrument Society of America, 1972.

1610. Thompson, R. W., R. L. Gilbreath, and F. Bielk. Alterations of porcine skin acid mucopolysaccharides in zinc deficiency. J. Nutr. 105:154-160, 1975.

1611. Thorne, D. W., and F. B. Wann. Nutrient Deficiencies in Utah Orchards. Utah Agricultural Experiment Station Bulletin 338. Logan: Utah State Agricultural College, 1950. 29 pp.

1612. Thorne, W. Zinc deficiency and its control. Adv. Agron. 9:31-65, 1957.

1613. Thorslund, A., and S. Linkskog. Studies of the esterase activity and the anion inhibition of bovine zinc and cobalt carbonic anhydrase. Eur. J. Biochem. 3:117-123, 1967.

1614. Thyresson, N. Acrodermatitis enteropathica. Report of a case healed with zinc therapy. Acta Derm. Venereol. 54:383-385, 1974.

1615. Tierney, D. P. Some Aspects of the Ecology of Naturally Occurring Populations of Submerged Vascular Hydrophytes in Municipal Wastewater Lagoons. III. Potential Use as a Feedstuff and a Soil Conditioner. Ph.D. Thesis. East Lansing: Michigan State University, 1972. 20 pp.

1616. Tietz, N. W., E. F. Hirsch, and B. Neyman. Spectrographic study of trace elements in cancerous and noncancerous human tissues. J.A.M.A. 165:2187-2192, 1957.

1617. Tiffin, L. O. Translocation of manganese, iron, cobalt, and zinc in tomato. Plant Physiol. 42:1427-1432, 1967.

1618. Tiffin, L. O. Translocation of micronutrients in plants, pp. 199-229. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Shoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

1619. Timperley, M. H., R. R. Brooks, and P. J. Peterson. The significance of essential and non-essential elements in plants in relation to biogeochemical prospecting. J. Appl. Ecol. 7:429-439, 1970.

1620. Tipton, I. H., and M. J. Cook. Trace elements in human tissue. Part II. Adult subjects from the United States. Health Phys. 9:103-145, 1963.

1621. Tipton, I. H., H. A. Schroeder, H. M. Perry, Jr., and M. J. Cook. Trace elements in human tissue. Part III. Subjects from Africa, the near and far East and Europe. Health Phys. 11:403-451, 1965.

1622. Titani, K., M. A. Hermodson, L. H. Ericsson, K. A. Walsh, and H. Neurath. Amino-acid sequence of thermolysin. Nature New Biol. 238:35-37, 1972.

1623. Tiwari, R. C., and B. M. Kumar. A common chemical extractant for estimating plant-available zinc in different soil types (peaty, red and alluvial). Plant Soil 41:689-693, 1974.

1624. Tobin, A. J. Carbonic anhydrase from parsley leaves. J. Biol. Chem. 245: 2656-2666, 1970.

1625. Topping, G. Concentrations of Mn, Co, Cu, Fe and Zn in the northern Indian Ocean and Arabian Sea. J. Marine Res. 27:318-326, 1969.

1626. Topping, G. Heavy metals in shellfish from Scottish waters. Aquaculture 1:379-384, 1973.

1627. Tourtelot, H. A. Minor-element composition and organic carbon content of marine and nonmarine shales of Late Cretaceous age in the western interior of the United States. Geochim. Cosmochim. Acta 28:1579-1604, 1964.

1628. Trentini, G. P., C. Ferrari de Gaetani, and M. S. Saviano. Modificazioni istomorfologiche ed istoenzimatiche del corticosurrene di ratto in carenza ed in trattamento con zinco. Boll. Soc. Ital. Biol. Sper. 45:607-610, 1969.

1629. Trubowitz, S., D. Feldman, S. W. Morgenstern, and V. M. Hunt. The isolation, purification and some properties of the alkaline phosphatase of human leucocytes. Biochem. J. 80:369-374, 1961.

1630. Tsui, C. The role of zinc in auxin synthesis in the tomato plant. Amer. J. Bot. 35:172-179, 1948.

1631. Turner, J. A., and L. R. Thompson. Health Hazards of Brass Foundries. I. Field Investigations of the Health Hazards of the Brass-Foundry Industry. II. Laboratory Studies Relating to the Pathology of Brass Foundrymen's Ague. U. S. Public Health Bulletin No. 157. Washington, D. C.: Government Printing Office, 1926. 75 pp.

1632. Turner, M. A. Effect of cadmium treatment on cadmium and zinc uptake by selected vegetable species. J. Environ. Qual. 2:118-119, 1973.

1633. Turner, R. G. The subcellular distribution of zinc and copper within the roots of metal-tolerant clones of Agrostis tenuis Sibth. New Phytol. 69:725-731, 1970.

1634. Turner, R. G., and R. P. G. Gregory. The use of radioisotopes to investigate heavy metal tolerance in plants, pp. 493-509. In Isotopes in Plant Nutrition and Physiology. Proceedings of a Symposium, Vienna, 5-9 Sept. 1966. Jointly Organized by the IAEA and FAO. Vienna: International Atomic Energy Agency, 1967.

1635. Tsuru, D., J. D. McConn, and K. T. Yasunobu. B. subtilis neutral protease, a zinc enzyme of high activity. Biochem. Biophys. Res. Commun. 15: 367-371, 1964.

1636. Tukey, H. B., Jr., H. B. Tukey, and S. H. Wittwer. Loss of nutrients by foliar leaching as determined by radioisotopes. Proc. Amer. Soc. Hort. Sci. 71:496-506, 1958.

1637. Tyler, A. Prolongation of life-span of sea urchin spermatozoa, and improvement of the fertilization-reaction, by treatment of spermatozoa and eggs with metal-chelating agents (amino acids, versene, dedtc, oxine, cupron). Biol. Bull. 104:224-239, 1953.

1638. Tyler, G. Moss analysis -- a method for surveying heavy metal deposition, pp. 129-132. In H. M. Englund, and W. T. Beery, Eds. Proceedings of the Second International Clean Air Congress. Held at Washington, D. C., Dec. 6-11, 1970. New York: Academic Press, 1971.

1639. Ullrey, D. E., W. T. Ely, and R. L. Covert. Iron, zinc and copper in mare's milk. J. Anim. Sci. 38:1276-1277, 1974.

1640. Underwood, E. J. Zinc, pp. 208-252. In Trace Elements in Human and Animal Nutrition. (3rd ed.) New York: Academic Press, 1971.

1641. Underwood, E. J. Zinc toxicity, p. 243. In Trace Elements in Human and Animal Nutrition. (3rd. ed.) New York: Academic Press, 1971.

1642. Underwood, E. J., and M. Somers. Studies of zinc nutrition in sheep. I. The relation of zinc to growth, testicular development, and spermatogenesis in young rams. Austral. J. Agric. Res. 20:889-897, 1969.

1643. U. S. Environmental Protection Agency. Office of Water and Hazardous Materials. Effluent Guidelines Division. Development Document for Interim Final Effluent Limitations Guidelines and Proposed New Source Performance Standards for the Zinc Segment of the Nonferrous Metals Manufacturing Point Source Category. EPA 440/1-75/032. Washington, D. C.: U. S. Environmental Protection Agency, 1975. 155 pp.

1643a. U. S. Atomic Energy Commission. Part 20--Standards for protection against radiation, pp. 161-188. In Codes of Federal Regualtions. 10. Energy. Parts 0 to 199. Revised as of January 1, 1975. Washington, D. C.: U. S. Government Printing Office, 1975.

1644. Harley, J. H., Ed. HASL Procedures Manual. HASL-300. (Present Edition 1972, Revisions Supplied Annually) New York: U. S. Energy Research & Development Administration, Health and Safety Laboratory, 1976. pp. B-03-01--B-03-05, B-04-01--B-04-14, and E-00-04-01--E-00-04-03.

1644a. McMahon, A. D. J. H. Hague, and H. R. Babitzke. Zinc, pp. 1303-1343. In Bureau of Mines. Minerals Yearbook 1973. Vol. 1. Metals, Minerals and Fuels. Washington, D. C.: U. S. Government Printing Office, 1975.

1645. U. S. Bureau of Yards and Docks. Radioactivity in Water Supply and Waste Water Systems: Peacetime Detection and Control. Department of the Navy NAVDOCKS MO-218. Washington, D. C.: U. S. Government Printing Office, 1962. 77 pp.

1645a. U. S. Department of Health, Education, and Welfare. Public Health Service. Air Quality Data from the National Air Surveillance Networks and Contributing State and Local Networks. 1966 Edition. Air Quality and Emission Data Program. National Air Pollution Control Administration Publication No. APTD 68-9. Washington, D. C.: U. S. Government Printing Office, 1968. 157 pp.

1645b. U. S. Department of Health, Education, and Welfare. Zinc, pp. 55-56. In Public Health Service Drinking Water Standards. Revised 1962. Rockville, Md.: U. S. Department of Health, Education, and Welfare, 1962.

1646. U. S. Department of Health, Education, and Welfare. Community Water Supply Study. Analysis of National Survey Findings. Cincinnati: U. S. Department of Health, Education, and Welfare. Bureau of Water Hygiene, 1970. 111 pp.

1646a. U. S. Environmental Protection Agency. Helena Valley, Montana, Area Environmental Pollution Study. Office of Air Programs Publ. No. AP-91. Research Triangle Park, N. C.: U. S. Environmental Protection Agency, 1972. 179 pp.

1646b. U. S. Environmental Protection Agency. Chemical Analysis of Interstate Carrier Water Supply Systems. EPA-430/9-75-005. Washington, D. C.: U. S. Government Printing Office, 1975. 90 pp.

1647. United States Pharmacopeia. Nineteenth Revision. Offical from July 1, 1975. Rockville, Md.: United States Pharmacopeial Convention, Inc., 1975. 824 pp.

1648. Uthe, J. F., and E. G. Bligh. Preliminary survey of heavy metal contamination of Canadian freshwater fish. J. Fish. Res. Board Can. 28:786-788, 1971.

1649. Vachek, H., and J. L. Wood. Purification and properties of mercaptopyruvate sulfur transferase of Escherichia coli. Biochim. Biophys. Acta 258:133-146, 1972.

1650. Valberg, L. S., J. M. Holt, and R. T. Card. Erythrocyte magnesium, copper and zinc in malignant disease affecting the hemopoietic system. Cancer 19:1833-1841, 1966.

1651. Valberg, L. S., J. M. Holt, and J. Szivek. Determination of calcium, magnesium, copper and zinc in red blood cells by emission spectrometry. Anal. Chem. 36:790-792, 1964.

1652. Vallee, B. L. Biochemistry, physiology and pathology of zinc. Physiol. Rev. 39:443-490, 1959.

1653. Vallee, B. L. The metabolic role of zinc. J.A.M.A. 162:1053-1057, 1956.

1654. Vallee, B. L. Zinc and its biological significance. A.M.A. Arch. Ind. Health 16:147-154, 1957.

1655. Vallee, B. L. Zinc Biochemistry, Physiology, Toxicology and Pathology. Report Prepared for the International Lead Zinc Research Organization, Inc., 1968. 103 pp.

1655a. Vallee, B. L., and M. D. Altschule. Zinc in the mammalian organism, with particular reference to carbonic anhydrase. Physiol. Rev. 29:370-388, 1949.

1656. Vallee, B. L., and J. G. Gibson, 2nd. An improved dithizone method for the determination of small quantities of zinc in blood and tissue samples. J. Biol. Chem. 176:435-443, 1948.

1657. Vallee, B. L., and H. Neurath. Carboxypeptidase, a zinc metalloenzyme. J. Biol. Chem. 217:253-262, 1955.

1658. Vallee, B. L., and F. L. Hoch. Zinc, a component of yeast alcohol dehydrogenase. Proc. Nat. Acad. Sci. U.S.A. 41:327-338, 1955.

1659. Vallee, B. L., and F. L. Hoch. Zinc in horse liver alcohol dehydrogenase. J. Biol. Chem. 225:185-196, 1957.

1660. Vallee, B. L., F. L. Hoch, and W. L. Hughes, Jr. Studies on metalloproteins. Soluble zinc-containing protein extracted from human leucocytes. Arch. Biochem. Biophys. 48:347-360, 1954.

1661. Vallee, B. L., and H. Neurath. Carboxypeptidase, a zinc metalloprotein. J. Amer. Chem. Soc. 76:5006, 1954. (letter)

1662. Vallee, B. L., E. A. Stein, W. N. Sumerwell, and E. H. Fischer. Metal content of α-amylases of various origins. J. Biol. Chem. 234: 2901-2905, 1959.

1663. Vallee, B. L., and D. D. Ulmer. Biochemical effects of mercury, cadmium and lead. Ann. Rev. Biochem. 41:91-128, 1972.

1664. Vallee, B. L., and W. E. C. Wacker. Metalloproteins. In H. Neurath, Ed. The Proteins. Composition, Structure, and Function. (2nd ed.) Vol. 5. New York: Academic Press, 1970. 192 pp.

1665. Vallee, B. L., and W. E. C. Wacker. Zinc, a component of rabbit muscle lactic dehydrogenase. J. Amer. Chem. Soc. 78:1771-1772, 1956. (letter)

1666. Vallee, B. L., W. E. C. Wacker, A. F. Bartholomay, and F. L. Hoch. Zinc metabolism in hepatic dysfunction. Ann. Intern. Med. 50:1077-1091, 1959.

1667. Vallee, B. L., W. E. C. Wacker, A. F. Bartholomay, and F. L. Hoch. Zinc metabolism in hepatic dysfunction. II. Correlation of metallic patterns with biochemical findings. New Engl. J. Med. 257:1055-1065, 1957.

1668. Vallee, B. L., W. E. C. Wacker, A. F. Bartholomay, and E. D. Robin. Zinc metabolism in hepatic dysfunction. I. Serum zinc concentrations in Laennec's cirrhosis and their validation by sequential analysis. New Engl. J. Med. 255:403-408, 1956.

1668a. van Adrichem, P. W. M., J. M. van Leeuwen, and J. J. van Kluijve. Parakeratosis of the skin in calves. Tids. Diergeneeskd. 95:1170-1176, 1970. (in Dutch, summary in English)

1669. van Campen, D. Competition between copper and zinc during absorption, pp. 287-298. In C. F. Mills, Ed. Trace Element Metabolism in Animals. Proceedings of WAAP/IBP International Symposium, Aberdeen, Scotland, July 1969. London: E. & S. Livingston, 1970.

1670. van Campen, D. R. Effects of zinc, cadmium, silver and mercury on the absorption and distribution of copper-64 in rats. J. Nutr. 88:125-130, 1966.

1671. van Campen, D. R., and W. A. House. Effect of a low protein diet on retention of an oral dose of ^{65}Zn and on tissue concentrations of zinc, iron and copper in rats. J. Nutr. 104:84-90, 1974.

1672. Van Campen, D. R., and P. U. Scaife. Zinc interference with copper absorption in rats. J. Nutr. 91:473-476, 1967.

1673. van Lear, D. H., and W. H. Smith. Relationships between macro- and micronutrient nutrition on slash pine on three coastal plain soils. Plant Soil 36:331-347, 1972.

1674. van Weers, A. Uptake and loss of ^{65}Zn and ^{60}Co by the mussel Mytilus edulis L., pp. 385-401. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA in Seattle, U.S.A. 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

1675. Vance, P. G., B. B. Keele, Jr., and K. V. Rajagopalan. Superoxide dismutase from Streptococcus mutans. Isolation and characterization of two forms of the enzyme. J. Biol. Chem. 247:4782-4786, 1972.

1676. Vandegrift, A. E., L. J. Shannon, E. W. Lawless, P. G. Gorman, E. E. Sallee, and M. Reichel. Emission sources and rates from primary zinc smelting, pp. 277-281. In Particulate Pollutant System Study. Volume III. Handbook of Emission Properties. APTD-0745. Kansas City, Missouri: Midwest Research Institute, 1971.

1677. Vander, A. J. Effects of zinc, cadmium and mercury on renal transport systems. Amer. J. Physiol. 204:781-784, 1963.

1677a. van der Bas, J. M., and H. Mulder. The zinc content of cows' milk. Neth. Milk Dairy J. 18:103-107, 1964.

1678. van der Borght, O., and S. van Puymbroeck. Ionic regulation and radiocontamination: "Active transport" of alkaline earth ions in freshwater gastropods (Mollusca), pp. 925-930. In B. Åberg and F. P. Hungate, Eds. Radioecological Concentration Processes. Proceedings of an International Symposium held in Stockholm 25-29 April, 1966. New York: Pergamon, 1967.

1679. Vanderploeg, H. A. Rate of zinc uptake by Dover sole in the northeast Pacific Ocean: Preliminary model and analysis, pp. 840-848. In D. J. Nelson, Ed. Radionuclides in Ecosystems. Vol. 2. Proceedings of the Third National Symposium on Radioecology, May 10-12, 1971. CONF-710501. Oak Ridge: U. S. Atomic Energy Commission, (not dated).

1680. Vanderploeg, H. A. The Dynamics of ^{65}Zn in Benthic Fishes and Their Prey off Oregon. Ph.D. Thesis. Corvallis: Oregon State University, 1973. 112 pp.

1680a. van Peenen, H. J., and F. V. Lucas. Zinc in liver disease. Arch. Path. 72:700-702, 1961.

1680b. van Peenen, H. J., and A. Patel. Tissue zinc and calcium in chronic disease. Arch. Path. 77:53-56, 1961.

1681. van Raaphorst, J. G., A. W. van Weers, and H. M. Haremaker. Loss of zinc and cobalt during dry ashing of biological material. Analyst 99: 523-527, 1974.

1682. Van Reen, R. Effects of excessive dietary zinc in the rat and interrelationship with copper. Arch. Biochem. Biophys. 46:337-344, 1953.

1683. van Rensburg, S. J. Failure of low protein and zinc intakes to influence nitrosamine-induced oesophageal carcinogenesis. S. Afr. Med. J. 46: 1137-1138, 1972.

1684. Verpoorte, J. A., S. Mehta, and J. T. Edsall. Esterase activities of human carbonic anhydrases B and C. J. Biol. Chem. 242:4221-4229, 1967.

1685. Verrilli, R. A., L. W. Brady, M. N. Croll, W. C. Hunsicker, and R. C. Uhlman. Zn-65 uptake by the prostate. J. Urol. 88:664-666, 1962.

1686. Viets, F. G., Jr. Zinc deficiency in the soil-plant system, pp. 90-128. In A. S. Prasad, Ed. Zinc Metabolism. Springfield, Ill.: Charles C Thomas, 1966.

1687. Viets, F. G., Jr., L. C. Boawn, and C. L. Crawford. The effect of nitrogen and type of nitrogen carrier on plant uptake of indigenous and applied zinc. Soil Sci. Soc. Amer. Proc. 21:197-201, 1957.

1688. Viets, F. G., Jr., L. C. Boawn, and C. L. Crawford. Zinc content of bean plants in relation to deficiency symptons and yield. Plant Physiol. 29:76-79, 1954.

1689. Viets, F. G., Jr., L. C. Boawn, and C. L. Crawford. Zinc contents and deficiency symptoms of 26 crops grown on a zinc-deficient soil. Soil Sci. 78:305-316, 1954.

1690. Viets, F. G., Jr., L. C. Boawn, C. L. Crawford, and C. E. Nelson. Zinc deficiency in corn in Central Washington. Agron. J. 45:559-565, 1953.

1691. Vigliani, E. C. The biopathology of cadmium. Amer. Ind. Hyg. Assoc. J. 30:329-340, 1969.

1692. Vikbladh, I. Studies on zinc in blood. II. Scand. J. Clin. Lab. Invest. 3(Suppl. 2):1-74, 1951.

1693. Vinande, R., B. Knezek, J. Davis, E. Doll, and J. Melton. Field and laboratory studies with zinc and iron fertilization of pea beans, corn, and potatoes in 1967. Mich. Agric. Exp. Stat. Q. Bull. 50:625-638, 1968.

1694. Vine, J. D., and E. B. Tourtelot. Geochemistry of black shale deposits -- a summary report. Econ. Geol. 65:253-272, 1970.

1695. Vinogradov, A. P. The Elementary Chemical Composition of Marine Organisms. Sears Foundation for Marine Research Memoir II. (translated from Russian by J. Efron and J. K. Setlow) New Haven: Yale University, 1953. 647 pp.

1696. Vinogradov, A. P. The Geochemistry of Rare and Dispersed Chemical Elements in Soils. (2nd ed.) New York: Consultants Bureau, Inc., 1959. 209 pp. (translated from Russian)

1697. Vogel, J., and J. Deshusses. Sur la teneur des vins en zinc. Mitteil. Gebiete Lebensmittel. Hyg. 53:269-271, 1962.

1698. Vogels, G. D., and C. Van der Drift. Allantoinases from bacterial, plant and animal sources. II. Effect of bivalent cations and reducing substances on the enzymic activity. Biochim. Biophys. Acta 122: 497-509, 1966.

1699. Vohra, P. A review of nutrition of Japanese quail. World Poult. Sci. J. 27:26-34, 1971.

1699a. Vohra, P., and J. R. Heil. Selection of zinc-supplemented diets by turkey poults. Poult. Sci. 48:1118-1120, 1969.

1700. Vohra, P., and F. H. Kratzer. Zinc, copper and manganese toxicities in turkey poults and their alleviation by EDTA. Poult. Sci. 47:699-704, 1968.

1701. Voigt, G. E., and T. Saldeen. Über den Schutzeffekt des Zinks gegenüber mangansulfat- oder Kohlenstofftetrachlorid-induzierten Leberschäden. Frankfurter Z. Path. 74:572-578, 1965.

1702. Volini, M., F. DeToma, and J. Westley. Dimeric structure and zinc content of bovine liver rhodanese. J. Biol. Chem. 242:5220-5225, 1967.

1703. Volkov, N. F. Cobalt, manganese and zinc content in the blood and internal organs of patients with atherosclerosis. Ter. Arkh. 34(12):52-56, 1962. (in Russian, summary in English)

1704. Volkovic, V., D. Miljanic, R. M. Wheeler, R. B. Liebert, T. Zabel, and G. C. Phillips. Variation in trace metal concentrations along single hairs as measured by proton-induced x-ray emission photometry. Nature 243:543-544, 1973.

1705. von Lehmden, D. J., R. H. Jungers, and R. E. Lee, Jr. Determination of trace elements in coal, fly ash, fuel oil, and gasoline -- A preliminary comparison of selected analytical techniques. Anal. Chem. 46: 239-245, 1974.

1706. von Wartburg, J.-P., J. L. Bethune, and B. L. Vallee. Human liver-alcohol dehydrogenase. Kinetic and physicochemical properties. Biochemistry 3:1775-1782, 1964.

1707. Vorobieva, A. I. Copper and zinc balances in the organism of children aged 2, 6, and 8-10 years. Vopr. Pitan. 26(4):28-30, 1967. (in Russian, summary in English)

1708. Vorobieva, A. I., and N. A. Bolshanina. The content of zinc and iron in the food rations of inmates of Tomsk Children's institutions. Vopr. Pitan. 23(5):78-79, 1964.

1709. Voth, J. L. Spectrographic method for determination of trace elements in milk. Anal. Chem. 35:1957-1958, 1963.

1710. Wacker, W. E. C. Nucleic acids and metals. III. Changes in nucleic acid, protein, and metal content as a consequence of zinc deficiency in Euglena gracilis. Biochemistry 1:859-865, 1962.

1711. Wacker, W. E. C., D. D. Ulmer, and B. L. Vallee. Metalloenzymes and myocardial infarction. II. Malic and lactic dehydrogenase activities and zinc concentrations in serum. New Engl. J. Med. 255:449-456, 1956.

1712. Wacker, W. E. C., and B. L. Vallee. Nucleic acids and metals. I. Chromium, manganese, nickel, iron, and other metals in ribonucleic acid from diverse biological sources. J. Biol. Chem. 234:3257-3262, 1959.

1713. Wagner, F. W., and J. M. Prescott. Purification and some characteristics of a snake venom proteinase. Fed. Proc. 25:590, 1966. (abstract)

1714. Waisel, Y., and Z. Shapira. Functions performed by roots of some submerged hydrophytes. Israeli J. Bot. 20:69-77, 1971.

1715. Wakeley, J. C. N., B. Moffatt, A. Crook, and J. R. Mallard. The distribution and radiation dosimetry of zinc-65 in the rat. Int. J. Appl. Rad. Isot. 7:225-232, 1960.

1715a. Walker, B. E., J. B. Dawson, J. Kelleher, and M. S. Losowsky. Plasma and urinary zinc in patients with malabsorption syndromes or hepatic cirrhosis. Gut 14:943-948, 1973.

1716. Wallace, A., A. ElGazzar, and G. V. Alexander. High phosphorous levels on zinc and other heavy metal concentrations in Hawkeye and PI54619-5-1 soybeans. Commun. Soil Sci. Plant Anal. 4:343-345, 1973.

1717. Wallace, A., and R. T. Mueller, Responses of plants to zinc and manganese chelates. Soil Sci. Soc. Amer. Proc. 23:79, 1958.

1718. Wallace, A., and E. M. Romney. The effect of zinc sources on micronutrient contents of Golden Cross Bantam corn. Soil Sci. 109:66-67, 1970.

1719. Wallace, A., E. M. Romney, V. Q. Hale, and R. M. Hoover. Effect of soil temperature and zinc application on yields and micronutrient content of four crop species grown together in a glasshouse. Agron. J. 61: 567-568, 1969.

1720. Waller, W. T., M. L. Dahlberg, R. E. Sparks, and J. Cairns, Jr. A computer simulation of the effects of superimposed mortality due to pollutants on populations of fathead minnows (Pimephales promelas). J. Fish. Res. Board Can. 28:1107-1112, 1971.

1721. Wallihan, E. F., and L. Heymann-Herschberg. Some factors affecting absorption and translocation of zinc in citrus plants. Plant Physiol. 31:294-299, 1956.

1722. Walravens, P. A., and K. M. Hambidge. Growth of infants fed a zn-supplemented milk formula. Pediatr. Res. 9:310, 1975. (abstract)

1723. Walravens, P. A., K. M. Hambridge, M. L. Roth, S. B. White, and M. L. Anthony. Zinc nutritional status of head start children. Ped. Res. 9:300, 1975. (abstract)

1723a. Walshe, J. M. Disturbances of aminoacid metabolism following liver injury. A study by means of paper chromatography. Q. J. Med. 22:483-505, 1953.

1724. Walsh, L. M., and J. D. Beaton, Eds. Soil Testing and Plant Analysis. Madison, Wis.: Soil Science Society of America, Inc., 1973. 491 pp.

1725. Walsh, L. M., D. R. Steevens, H. D. Seibel, and G. G. Weis. Effect of high rates of zinc on several crops grown on an irrigated Plainfield sand. Commun. Soil Sci. Plant Anal. 3:187-195, 1972.

1726. Walters, M., and F. J. C. Roe. A study of the effects of zinc and tin administered orally to mice over a prolonged period. Food Cosmet. Toxicol. 3:271-276, 1965.

1727. Waltner, K. Über die Wirkung einiger Metalle. Naunyn Schmiedeberg's Arch. Exp. Path. Pharmakol. 141:123-128, 1929.

1728. Wang, K.-M. Zn(II)-activated acid phosphatase in liver and metanephros of developing chick. Experientia 24:424-425, 1968.

1729. Ward, R. L., and J. A. Happe. ^{35}Cl NMR studies of zinc adenosine diphosphate complexes. Biochem. Biophys. Res. Commun. 28:785-790, 1967.

1730. Warkany, J. Problems in applying teratologic observations in animals and man. Pediatrics 53:820, 1974.

1731. Warkany, J., and H. G. Petering. Congenital malformations of the brain produced by short zinc deficiencies in rats. Amer. J. Ment. Defic. 77:645-653, 1973.

1732. Warkany, J., and H. G. Petering. Congenital malformations of the central nervous system in rats produced by maternal zinc deficiency. Teratology 5:319-334, 1972.

1733. Warncke, D. D., and S. A. Barber. Diffusion of zinc in soil: I. The influence of soil moisture. Soil Sci. Soc. Amer. Proc. 36:39-42, 1972.

1734. Warncke, D. D., and S. A. Barber. Diffusion of zinc in soils: III. Relation to zinc adsorption isotherms. Soil Sci. Soc. Amer. Proc. 37:355-358, 1973.

1735. Warncke, D. D., and S. A. Barber. Diffusion of zinc in soil: II. The influence of soil bulk density and its interaction with soil moisture. Soil Sci. Soc. Amer. Proc. 36:42-46, 1972.

1736. Warnick, S. L., and H. L. Bell. The acute toxicity of some heavy metals to different species of aquatic insects. J. Water Pollut. Control Fed. 41:280-284, 1969.

1737. Warnock, R. E. Micronutrient uptake and mobility within corn plants (Zea mays L.) in relation to phosphorus-induced zinc deficiency. Soil Sci. Soc. Amer. Proc. 34:765-769, 1970.

1737a. Reed, S. C., et al. Wastewater Management by Disposal on the Land. Cold Regions Research and Engineering Laboratory Special Report 171. Hanover, N. H.: U. S. Army Corps of Engineers, 1972. 190 pp.

1737b. Waters, M. D., R. D. Moore, J. J. Amato, and J. C. Houck. Zinc sulfate-failure as an accelerator of collagen biosynthesis and fibroblast proliferan. Proc. Soc. Exp. Biol. Med. 138:373-377, 1971.

1738. Watson, D. G., J. J. Davis, and W. C. Hanson. Zinc-65 in marine organisms along the Oregon and Washington coasts. Science 133:1826-1828, 1961.

1738a. Watson, C. J., and S. Schwartz. The excretion of zinc uroporphyrin in idopathic porphyria. J. Clin. Invest. 20:440-441, 1941. (abstract)

1739. Waygood, E. R., and K. A. Clendenning. Carbonic anhydrase in green plants. Can. J. Res. 28C:673-689, 1950.

1740. Weast, R. C., Ed. CRC Handbook of Chemistry and Physics. (56th ed.) Cleveland, Ohio: The Chemical Rubber Co., 1975. [paged by sections]

1741. Webb, M. Protection by zinc against cadmium toxicity. Biochem. Pharmacol. 21:2767-2771, 1972.

1742. Webb, M. The biological action of cobalt and other metals. IV. Inhibition of α-oxoglutarate dehydrogenase. Biochim. Biophys. Acta 89: 431-446, 1964.

1743. Webley, D. H., R. B. Duff, and G. Anderson. Metabolism of iron-, zinc-manganese-deficient Nocardia opaca. J. Gen. Microbiol. 29:179-187, 1962.

1743a. Wedepohl, K. H. Zinc, Chapter 30, Sections B-O. In Handbook of Geochemistry. Vol. II-3. New York: Springer-Verlag, 1972.

1744. Wegener, W. S., and A. H. Romano. Zinc stimulation of RNA and protein synthesis in Rhizopus nigricans. Science 142:1669-1670, 1963.

1744a. Wegger, I., and B. Palludan. Zinc metabolism in swine. IV. B. Resorption and distribution of Zn^{65} in normal and zinc deficient newborn pigs. Aarsberetn. Inst. Sterilitetsforstk. Kgl. Vet. Landbohoejsk. 16: 51-73, 1973. (in Danish, summary in English)

1745. Wegner, T. N., D. E. Ray, C. D. Lox, and G. H. Stott. Effect of stress on serum zinc and plasma corticoids in dairy cattle. J. Dairy Sci. 56: 748-752, 1973.

1746. Weil, L., T. S. Scibles, and F. T. Herskovits. Photooxidation of bovine insulin sensitized by methylene blue. Arch. Biochem. Biophys. 111: 308-320, 1965.

1747. Weinberg, E. D. Biosynthesis of secondary metabolites: Roles of trace metals. Adv. Microb. Physiol. 4:1-44, 1970.

1748. Weinberg, E. D. Iron and susceptibility to infectious disease. Science 184:952-956, 1974.

1748a. Wiesel, L. L. Metal chelation in the mechanism of action of glucogenic corticosteroids. Metabolism 8:256-264, 1959.

1749. Weiss, R. H. Visible spectrophotometry, pp. 557-573. In M. Zief and R. Speights, Eds. Ultrapurity: Methods and Techniques. New York: Marcel Dekker, Inc., 1972.

1750. Weissman, N., and V. J. Pileggi. Zinc, pp. 703-707. In R. J. Henry, D. C. Cannon, and J. W. Winkelman, Eds. Clinical Chemistry. Principles and Technics. (2nd ed.) New York: Harper & Row, 1974.

1751. Weitzel, G., E. Buddecke, A.-M. Fretzdorff, F.-J. Strecker, and U. Roester. Zink im Augenhintergrund des Seehundes. Hoppe-Seyler's Z. Physiol. Chem. 304:1-10, 1956.

1752. Weitzel, G., and A.-M. Fretzdorff. Zink in den Augen von Säugetieren. Hoppe-Seyler's Z. Physiol. Chem. 292:221-231, 1953.

1753. Weitzel, G., and A.-M. Fretzdorff. Zinkbestimmung in biologischem Material. Hoppe-Seyler's Z. Physiol. Chem. 292:212-221, 1953.

1754. Weitzel, G., F.-J. Strecker, U. Roester, E. Buddecke, and A.-M. Fretzdorff. Zink im Tapetum lucidum. Hoppe-Seyler's Z. Physiol. Chem. 296:19-30, 1954.

1754a. Weitzel, G., E. Buddecke, A.-M. Fretzdorff, F.-J. Strecker, and U. Roester. Struktur der im Tapetum lucidum von Hund und Fuchs enthaltenen Zink-verbindung. Hoppe-Seyler's Z. Physiol. Chem. 299:193-213, 1955.

1755. Welch, R. M., W. A. House, and W. H. Allaway. Availability of zinc from pea seed to rats. J. Nutr. 104:733-740, 1974.

1755a. Wells, B. T., and R. K. Winkelmann. Acrodermatitis enteropathica. Arch. Derm. 84:40-52, 1961.

1756. Weser, U., R. Prinz, A. Schallies, A. Fretzdorff, P. Krauss, W. Voelter, and W. Voetsch. Microbial and hepatic cuprein (superoxide dismutase). Hoppe-Seyler's Z. Physiol. Chem. 353:1821-1831, 1972.

1757. Weser, U., S. Seeber, and P. Warnecke. Reactivity of Zn^{2+} on nuclear DNA and RNA biosynthesis of regenerating rat liver. Biochim. Biophys. Acta 179:422-428, 1969.

1757a. Weser, U., S. Seeber, and P. Warnecke. Zur Wirkung von Zn^{++} auf die Nukleinsäurebiosynthese von Aszites-Tumorzellen. Experientia 25: 489-490, 1969.

1758. West, P. W. Chemical analysis of inorganic particulate pollutants, pp. 147-185. In A. C. Stern, Ed. Air Pollution. Vol. 2. Analysis, Monitoring, and Surveying. (2nd ed.) New York: Academic Press, 1968.

1759. West, P. W., and S. K. Thabet. Microdetermination of zinc by means of reagent crayons and the ring-oven technique. Anal. Chim. Acta 37: 246-252, 1967.

1760. Westmoreland, N. Connective tissue alterations in zinc deficient rats. Fed. Proc. 30:1001-1010, 1971.

1761. Whanger, P. D., and P. H. Weswig. Effect of supplementary zinc on the intracellular distribution of hepatic copper in rats. J. Nutr. 101: 1093-1098, 1971.

1762. Wheatland, A. B., and B. J. Borne. Some changes in polluted water during percolation through soil. Water Waste Treat. J. 8:330-335, 1961.

1763. White, H. S. Inorganic elements in weighed diets of girls and young women. J. Amer. Diet. Assoc. 55:38-43, 1969.

1764. White, A., P. Handler, and E. L. Smith. Genetic aspects of metabolism, pp. 598-610. In Principles of Biochemistry. (3rd ed.) New York: McGraw-Hill Book Co., 1964.

1765. White, A., P. Handler, and E. L. Smith. Principles of Biochemistry. (3rd ed.) New York: McGraw-Hill Book Co., 1964. p. 821.

1766. White, I. G. The toxicity of heavy metals to mammalian spermatozoa. Austral. J. Exp. Biol. Med. Sci. 33:359-366, 1955.

1766a. Whitmore, W. F., Jr. Comments on zinc in the human and canine prostates. Nat. Cancer Inst. Monogr. 12:337-340, 1963.

1767. Whitney, P. L., G. Fölsch, P. O. Nyman, and B. G. Malmström. Inhibition of human erythrocyte carbonic anhydrase B by chloracetyl sulfonamides with labeling of the active site. J. Biol. Chem. 242:4206-4211, 1967.

1768. Whitney, P. L., P. O. Nyman, and B. G. Malmström. Inhibition and chemical modifications of human erythrocyte carbonic anhydrase B. J. Biol. Chem. 242:4212-4220, 1967.

1769. Whitton, B. A. Toxicity of zinc, copper and lead to Chlorophyta from flowing waters. Arch. Mikrobiol. 72:353-360, 1970.

1770. Wichmann, H. J. Isolation and determination of traces of metals. The dithizone system. Ind. Eng. Chem. Anal. Ed. 11:66-72, 1939.

1770a. Widdowson, E. M. Chemical analysis of the body, pp. 31-47. In J. Brožek, Ed. Human Body Composition: Approaches and Application. New York: Pergamon Press, 1965.

1771. Widdowson, E. M., H. Chan, G. E. Harrison, and R. D. G. Miller. Accumulation of Cu, Zn, Mn, Cr and Co in the human liver before birth. Biol. Neonate 20:360-367, 1972.

1771a. Widdowson, E. M., J. Dauncey, and J. C. L. Shaw. Trace elements in foetal and early postnatal development. Proc. Nutr. Soc. 33:275-284, 1974.

1772. Wiley, D. C., D. R. Evans, S. G. Warren, C. H. McMurray, B. F. P. Edwards, W. A. Franks, and W. N. Lipscomb. The 5.5 Å resolution structure of the regulatory enzyme, aspartate transcarbamylase. Cold Spring Harbor Symposium on Quantitative Biology 36:285-290, 1971.

1773. Wilkins, P. J., and I. E. Dreosti. Plasma zinc levels in zinc-deficient rats. Agrochemophysica 2:83-84, 1970.

1774. Wilkins, P. J., P. C. Grey, and I. E. Dreosti. Plasma zinc as an indicator of zinc status in rats. Brit. J. Nutr. 27:113-120, 1972.

1775. Wilkinson, H. F. Movement of micronutrients to plant roots, pp. 139-169. In J. J. Mortvedt, P. M. Giordano, and W. L. Lindsay, Eds. Micronutrients in Agriculture. Proceedings of a Symposium held at Muscle Schoals, Alabama, April 20-22, 1971. Madison, Wis.: Soil Science Society of America, 1972.

1776. Wilkinson, H. F., J. F. Loneragan, and J. P. Quirk. The movement of zinc to plant roots. Soil Sci. Soc. Amer. Proc. 32:831-833, 1968.

1777. Williams, C. H., and C. W. E. Moore. The effect of stage of growth on the copper, zinc, manganese, and molybdenum contents of Algerian oats grown on thirteen soils. Austral. J. Agric. Res. 3:343-361, 1952.

1778. Williams, R. B. Intestinal alkaline phosphatase and inorganic pyrophosphatase activities in the zinc-deficient rat. Brit. J. Nutr. 27:121-130, 1972.

1779. Williams, R. B., and J. K. Chesters. The effects of early zinc deficiency on DNA and protein synthesis in the rat. Brit. J. Nutr. 24:1053-1059, 1970.

1780. Williams, R. B., P. Demertzis, and C. F. Mills. The effects of dietary zinc concentration on reproduction in the rat. Proc. Nutr. Soc. 32:3A-4A, 1973. (abstract)

1781. Williams, R. B., and C. F. Mills. Relationships between zinc deficiency and folic acid status of the rat. Proc. Nutr. Soc. 32:2A-3A, 1973. (abstract)

1782. Williams, R. B., and C. F. Mills. The experimental production of zinc deficiency in the rat. Brit. J. Nutr. 24:989-1003, 1970.

1783. Williams, R. B., C. F. Mills, J. Quarterman, and A. C. Dalgarno. Effect of zinc deficiency on the in vivo incorporation of ^{32}P into rat-liver nucleotides. Biochem. J. 95:29P, 1965. (abstract)

1784. Williams, R. H., S. A. Walsh, and J. W. Ensinck. Effects of metals upon the conversion of adenosine triphosphate to adenosine 3', 5'-monophosphate in lipocytes. Proc. Soc. Exp. Biol. Med. 128:279-283, 1968.

1785. Williams, R. O., and L. A. Loeb. Zinc requirement for DNA replication in stimulated human lymphocytes. J. Cell Biol. 58:594-601, 1973.

1786. Willoughby, R. A., E. MacDonald, B. J. McSherry, and G. Brown. Lead and zinc poisoning and the interaction between Pb and Zn poisoning in the foal. Can. J. Comp. Med. 36:348-359, 1972.

1787. Wilson, E. L., P. E. Burger, and E. B. Dowdle. Beef-liver 5-aminolevulinic acid dehydratase. Purification and properties. Eur. J. Biochem. 29:563-571, 1972.

1788. Winchester, J. W. Application of neutron activation analysis to the investigation of natural and pollution aerosols. J. Radioanal. Chem. 19:311-317, 1974.

1789. Winder, F., and J. M. Denneny. Effect of iron and zinc on nucleic acid and protein synthesis in Mycobacterium smegmatis. Nature 184:742-743, 1959.

1790. Winder, F. G., and D. S. Barber. Effects of hydroxyurea, nalidixic acid and zinc limitation on DNA polymerase and ATP-dependent deoxyribonuclease activities of Mycobacterium smegmatis. J. Gen. Microbiol. 76: 189-196, 1973.

1790a. Winell, M. An international comparison of hygienic standards for chemicals in the work environment. Ambio 4:34-36, 1975.

1791. Wintersberger, E. Isolation and structure of an active-center peptide of bovine carboxypeptidase B containing the zinc-binding sulfhydryl group. Biochemistry 4:1533-1536, 1965.

1792. Wintersberger, E., D. J. Cox, and H. Neurath. Bovine pancreatic procarboxypeptidase B. I. Isolation, properties, and activation. Biochemistry 1:1069-1078, 1962.

1793. Wintersberger, E., H. Neurath, T. L. Coombs, and B. L. Vallee. A zinc-binding thiol group in the active center of the bovine carboxypeptidase B. Biochemistry 4:1526-1532, 1965.

1793a. Wisner, H. K., J. B. Lynch, D. L. Larson, and S. R. Lewis. Correction of retarded epidermal regeneration due to sulfamylon by administration of oral zinc, pp. 129-133. In P. Matter, T. L. Barclay, and Z. Konickova, Eds. Research in Burns. Transactions of the Third International Congress, held in Prague Sept. 20-25, 1970. Bern: Hans Huber Publishers, 1971.

1794. Withers, A. F. D., H. Baker, M. Musa, and T. L. Dormandy. Plasma-zinc in psoriasis. Lancet 2:278, 1968. (letter)

1795. Wittwer, S. H. Foliar absorption of plant nutrients. Adv. Front. Plant Sci. 8:161-182, 1964.

1796. Wixson, B. G., and E. Bolter. Evaluations of stream pollution and trace substances in the new lead belt of Missouri, pp. 143-152. In D. D. Hemphill, Ed. Trace Substances in Environmental Health - V. Proceedings of 5th Annual Conference of Trace Substance in Environmental Health held in Columbia, Missouri, June 29 - July 1, 1971. Columbia: University of Missouri, 1972.

1797. Wolfe, D. A. Levels of stable Zn and ^{65}Zn in Crassostrea virginica from North Carolina. J. Fish. Res. Board Can. 27:47-57, 1970.

1798. Wolfe, D. A. The estuarine ecosystem(s) at Beaufort, North Carolina. pp. 645-671. In L. E. Cronin, Ed. Estuarine Research. Vol. 1. Chemistry, Biology and the Estuarine System. New York: Academic Press, 1975

1799. Wolfe, D. A. The cycling of zinc in the Newport River estuary, North Carolina, pp. 79-99. In F. J. Vernberg and W. B. Vernberg, Eds. Pollution and Physiology of Marine Organisms. New York: Academic Press, 1974.

1800. Wolfe, D. A. Zinc enzymes in Crassostrea virginica. J. Fish. Res. Board Can. 27:59-69, 1970.

1801. Wolfe, D. A., F. A. Cross, and C. D. Jennings. The flux of Mn, Fe and Zn in an estuarine ecosystem, pp. 159-175. In Radioactive Contamination of the Marine Environment. Proceedings of a Symposium on the Interaction of Radioactive Contaminants with the Constituents of the Marine Environment held by IAEA in Seattle, U.S.A., 10-14 July 1972. Vienna: International Atomic Energy Agency, 1973.

1802. Wolff, H. Untersuchungen zur Pathophysiologie des Zinkstoffwechsels. Klin. Wochenschr. 34:409-418, 1956.

1802a. Delete 1802a--same as 1802

1803. Wood, J. G., and P. M. Sibly. Carbonic anhydrase activity in plants in relation to zinc content. Austral. J. Sci. Res. B 5:244-255, 1952.

1804. Woodbury, J., K. Lyons, R. Carretta, A. Hahn, and J. F. Sullivan. Cerebrospinal fluid and serum levels of magnesium, zinc, and calcium in man. Neurology 18:700-705, 1968.

1805. Woolsey, R. L., W. L. Anthony, and J. M. Hsu. Zinc deficiency and uridine-^{3}H incorporation into RNA in rat tissues. Fed. Proc. 33:700, 1974. (abstract)

1806. World Health Organization. Trace Elements in Human Nutrition. Report of a WHO Expert Committee. WHO Technical Report Series No. 532. Geneva:

1806a. World Health Organization. Zinc, p. 37. In European Standards for Drinking-Water. (2nd ed.) Geneva: World Health Organization, 1970.

1807. Wright, E. B., and T. L. Dormandy. Liver zinc in carcinoma. Nature 237: 166, 1972.

1808. Deleted.

1809. Wysocki, K., L. Owczarek, W. Fenrych, S. Gorski, and C. Majewski. The metabolism of radioactive zinc in the liver of rats damaged by a single administration of carbon tetrachloride. I. Radioisotopic studies. Acta Med. Polona 7:97-102, 1966.

1809a. Yamaguchi, T. Inhibition of glutamate dehydrogenase by bilirubin. J. Biochem. (Tokyo) 68:441-447, 1970.

1809b. Yatsozhinskiy, Yu. D., Ye. M. Bernikova, and V. N. Molotkov. Contents of microelements in lungs from patients with pulmonary tuberculosis. Vrach. Delo 1973(2):58-60. (in Russian)

1810. Yendt, E. R., and M. Cohanim. Ten years' experience with the use of thiazides in the prevention of kidney stones. Trans. Amer. Clin. Climatol. Assoc. 85:65-75, 1973.

1811. Yendt, E. R., G. F. Guay, and D. A. Garcia. The use of thiazides in the prevention of renal calculi. Can. Med. Assoc. J. 102:614-620, 1970.

1812. Yoshizumi, F. K., and J. E. Coleman. Metalloalkaline phosphatases from Bacillus subtilis: Physicochemical and enzymatic properties. Arch. Biochem. Biophys. 160:255-268, 1974.

1813. Yost, K. J., W. Bruns, J. E. Christian, F. M. Clikeman, R. B. Jacko, D. R. Masarik, W. W. McFee, A. W. McIntosh, J. E. Newman, R. I. Pietz, and A. M. Zimmer. The Environmental Flow of Cadmium and Other Trace Metals. Vol. I. Progress Report, July 1, 1972 to June 30, 1973. (Sponsored by National Science Foundation NSF (RANN) Grant GI-35106. Report NSF-RA-E73-016-A) West Lafayette, IN: Purdue University, 1973.

1814. Young, D. R., and T. R. Folsom. Loss of Zn^{65} from the California sea-mussel Mytilus californianus. Biol. Bull. 133:438-447, 1967.

1815. Young, E. G., and W. M. Langille. The occurrence of inorganic elements in marine algae of the Atlantic provinces of Canada. Can. J. Bot. 36:301-310, 1958.

1816. Yudovich, Ya. E., A. A. Korycheva, A. S. Obruchnikov, and Yu. V. Stepanov. Mean trace-element contents in coals. Geochem. Int. 9:712-720, 1972.

1816a. Zeman, F. J. Effect on the young rat of maternal protein restriction. J. Nutr. 93:167-173, 1967.

1816b. Zazgornik, J. Über die Zinkausscheidung im Schweiss und Harn bei chronischen Alkoholikern während Wärmeexposition. Res. Exp. Med. 160:252-254, 1973.

1816c. Zel'tser, M. E. The functional state of the thyroid gland in lead poisoning. Tr. Inst. Kraevoi. Patol. Akad. Nauk Kaz. SSR 10: 116-120, 1962. (in Russian)

1816d. Zazgornik, J., and P. Schmidt. Effects of zinc-containing dialysis membranes on zinc metabolism in patients on RDT, pp. 548-552. In J. S. Cameron, D. Fries and C. S. Ogg, Eds. Dialysis and Renal Transplantation. Proceedings of the Ninth Conference of the European Dialysis and Transplant Association, 1972. London: Sir Isaac Pitman and Sons, Ltd., 1972.

1817. Zeman, F. J., R. E. Shrader, and L. H. Allen. Persistent effects of maternal protein deficiency in postnatal rats. Nutr. Rep. Int. 7:421-436, 1973.

1817a. Zhukov, N. A., and T. S. Bakhina. Pathways and significance of redistribution of zinc in patients with chronic pancreatitis. Ter. Arkh. 44 (7):64-65, 1972. (in Russian, summary in English)

1818. Zief, M., and R. Speights, Eds. Ultrapurity: Methods and Techniques. New York: Marcel Dekker, Inc., 1972. 697 pp.

1819. Zielke, C. L., and C. H. Suelter. Rabbit muscle adenosine 5'-monophosphate aminohydrolase. Characterization as a zinc metalloenzyme. J. Biol. Chem. 246:2179-2186, 1971.

1820. Zinc and reproduction. Nutr. Rev. 27:16-18, 1969.

1821. Zinc in relation to DNA and RNA synthesis in regenerating rat liver. Nutr. Rev. 27:211-213, 1969.

1821a. Zubovic, P. Physicochemical properties of certian minor elements as controlling factors in their distribution in coal. Adv. Chem. 55:221-246, 1966.

1821b. Zipper, J., M. Medel, and R. Prager. Suppression of fertility by intrauterine copper and zinc in rabbits. Amer. J. Obstet. Gynecol. 105:529-534, 1969.

1822. Zubovic, P., N. B. Sheffey, and T. Stadnichenko. Distribution of Minor Elements in Some Coals in the Western and Southwestern Regions of the Interior Coal Province. A Study of 15 Minor Elements in Some of the Coals of Arkansas, Iowa, Missouri, Oklahoma and Texas. U. S. Geogical Survey Bulletin 1117-D. Washington, D. C.: U. S. Government Printing Office, 1967. 33 pp.

1823. Zubovic, P., T. Stadnichenko, and N. B. Sheffey. Distribution of Minor Elements in Coal Beds of the Eastern Interior Region. A Study of 15 Minor Elements in Coal Beds of Illinois, Indiana, and Western Kentucky. U. S. Geological Survey Bulletin No. 1117-B. Washington, D. C.: U. S. Government Printing Office, 1964. 41 pp.

1824. Zubovic, P., T. Stadnichenko, and N. B. Sheffey. Distribution of Minor Elements in Coals of the Appalachian Region. A Study of 15 Minor Elements in Some Coals in Ohio, Pennsylvania, Maryland, Kentucky, Tennessee, Alabama and Georgia. U. S. Geological Survey Bulletin 1117-C. Washington, D. C.: U. S. Government Printing Office, 1966. 37 pp.

1825. Zubovic, P., T. Stadnichenko, and N. B. Sheffey. Geochemistry of Minor Elements in Coals of the Northern Great Plains Coal Province. A Study of 15 Minor Elements in Some of the Coals of Montana, North Dakota, and Wyoming. U. S. Geological Survey 1117-A. Washington, D. C.: U. S. Government Printing Office, 1961. 58 pp.

1826. Zook, E. G., F. E. Greene, and E. R. Morris. Nutrient composition of selected wheats and wheat products. VI. Distribution of manganese, copper, nickel, zinc, magnesium, lead, tin, cadmium, chromium and selenium as determined by atomic absorption spectroscopy and colorimetry. Cereal Chem. 47:720-731, 1970.

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16. ABSTRACT

This report summarizes the available information on zinc as it relates to its effects on man and his environment. Zinc is found in most soils, but some areas are deficient in it. Metallurgic operations contribute to zinc contamination in air, water and soil. Trace amounts of zinc are essential for normal growth in plants, animals and humans, however, excessive levels can bring on zinc toxicosis. Zinc deficiency is known to have caused congenital malformations in pregnant rats. Severe liver disease is commonly associated with loss of total body zinc. Zinc is not a highly toxic substance. Zinc toxicosis may occur only when very high dose levels overwhelm the homeostatic mechanisms controlling zinc uptake and excretion. Reports suggest humans may ingest 500 mg to 1 g or more daily without adverse effects. Ten or more g taken as a single oral dose may produce gastrointestinal distress, including nausea, vomiting and diarrhea. There are also suggestions in the literature that even higher dosage may produce dizziness and perhaps increase blood levels of pancreatic enzymes. Inhalation of zinc has been related to metal fume fever, an acute disability of short duration that can occur when fume is inhaled from metal heated to a temperature above its melting point. With repeated exposure, some degree of tolerance may be built up, but it will be lost when exposure to fume ceases for a period as short as two days. The pathogenesis of this disorder, including the role of zinc in it, is not understood.

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