STUDY OF LEAD, COPPER, ZINC AND CADMIUM CONTAMINATION OF FOOD CHAINS OF MAN

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TABLE OF CONTENTS

		PAGE
I.	INTRODUCTION	2
II.	SUMMARY	5
III.	RECOMMENDATIONS	8
IV.	MATERIALS AND METHODS	11
	A. DESCRIPTION OF STUDY AREA	11
	B. SELECTION OF THE TEST FARM	13
	C. SELECTION OF THE CONTROL FARM	14
	D. DESCRIPTION OF THE TEST AND CONTROL COWS	17
	E. SAMPLE COLLECTION AND PREPARATION	19
	F. ATOMIC ABSORPTION SPECTROPHOTOMETRY PROCEDURES	25
	G. STATISTICAL PROCEDURES	26
V.	RESULTS AND DISCUSSION	33
	A. DUSTFALL	33
	B. SOIL	34
	C. VEGETATION - ROOTS	35
	D. VEGETATION - TOPS	35
	E. CATTLE HAIR	37
	F. CATTLE BLOOD	38
	G. MILK FROM COWS	39
	H. CATTLE TISSUES	76
	I. HUMAN CONSUMPTION OF MEAT AND MILK FROM COWS EXPOSED TO PRODUCTION SOURCES OF HEAVY METALS	76
	J. GARDEN CROPS	80
	K. LEAD TRANSLOCATION	80

PAGE

L. INTAKE AND ASSIMILATION OF LEAD BY CATTLE 87

M. METEOROLOGICAL EFFECTS AND SOURCES OF CONTAMINATION ON THE TEST FARM 94

VI. REFERENCES 103

VII. ACKNOWLEDGMENTS 108

VIII. APPENDIX TABLES 110

I.

INTRODUCTION

I. INTRODUCTION

Environmental contamination by cadmium (Cd), copper (Cu), lead (Pb) and zinc (Zn) involves several trophic levels of the ecosystem. It is well documented that airborne lead is translocated through food chains of soil, roots, foliage and domestic animals to man.[1] Lead is present in varying amounts in all natural foods. Cadmium is usually present in the environment in small amounts, but serious illnesses in human and domestic animal populations have been observed following ingestion of food and water contaminated by cadmium containing mining wastes.[2] Copper and zinc are considered essential elements for mammals and are translocated from the soil via various food chains.[3]

Under normal circumstances, the levels of these elements in the environment are determined by the geochemical composition of the region, and they are not high enough to adversely affect the health of indigenous animal or human populations. For example, the current soil lead concentrations in most rural areas are usually similar to the average content in the earth's crust, 10-15 ug/gm.[4] There is, however, evidence that there have been localized increases in lead concentrations in soil, and this is thought to be associated with an increased body burden of lead among certain population groups.

Lead ore mining and lead production results in the addition of Cd, Cu, Pb and Zn to the levels naturally present in the soil. The extent of this contamination depends upon the amounts contributed by various sources. In turn, the level of contamination will affect the quantities of these elements translocated in various food chains. Some of the food chains involve human foods, and environmental contaminants may be transported long distances in food products. Thus, remote human populations may be exposed via food products to contaminants originating in a mining area.

The primary purpose of this study was to investigate production sources of heavy metal contamination of food supplies. The objectives were:

1. estimation of annual contamination of soil in a lead mining area by Cd, Cu, Pb and Zn,

2. quantitation of these metals on vegetation,

3. estimation of intake by animals grazing on contaminated vegetation,

4. determination of levels of contamination of meat and milk produced by cattle grazing on contaminated pasture.

II.

SUMMARY

II. SUMMARY

A statistically designed study was conducted in the new lead producing region of southeastern Missouri to estimate the amount of soil, vegetation, meat and milk contamination by Cd, Cu, Pb and Zn. Dustfall, soil, root and vegetation tops were collected 4 times during a one year period at varying distances from the highway on a test farm exposed to lead production sources of heavy metal contamination and on a control farm outside the lead production area. Hair, blood, milk, liver, kidney cortex, diaphragm muscle and bone samples were collected for analysis by atomic absorption spectrophotometry from cows on the test and control farms. Between the first and last sampling, the lead concentration in soil at the 60 ft (18.3 m), 140 ft (42.7 m) and 220 ft (67.1 m) sites on the test farm increased 219%, 257% and 284%, respectively. The main sources of heavy metals at the test farm were stack emissions from a lead smelter, lead ore concentrate spillage from trucks, and dust from stockpiled ore at the smelter. High lead and cadmium dustfall and air filter samples at the test farm corresponded to wind conditions that would carry the smelter plume in the direction of the test farm, located approximately 800 meters from the stack. The results of analyses of dustfall and soil samples from the 3 sites at varying distances from the highway indicated that copper was a highway contaminant on the test farm but not on the control farm. The highest airborne, suspended Cd, Cu, Pb and Zn concentrations were observed in winter on the test farm. This corresponded with high dustfall, soil and vegetation levels and the time of greatest

increase in lead assimilation by a project owned test cow. There was a general dilution in the lead concentrations at each step of the food chain from soil to cattle tissues on the control farm, while on the test farm bio-magnification of lead in the grass roots was observed. This was attributed to higher airborne lead levels on the test farm, foliar absorption and deposition in the roots. On an equivalent basis, the test cow's blood Pb concentration at the end of the study period was 1/246 the test farm soil concentration and the control cow's blood Pb was 1/187 the control farm soil concentration. Analysis of cattle hair was a sensitive indicator of lead contamination; however, it had limited use in determining the body burden of lead because washed hair contains both exogenous and endogenous components. The liver, kidney, muscle and milk of the test cow contained very small amounts of cadmium, and the lead levels were 2.35 ug/gm, 3.75 ug/gm, 0.19 ug/gm and 13 ug/100 ml, respectively. The test cow milk Pb concentrations were 1.9 times that of the control cow, and Pb concentrations in milk from another cow exposed to only lead ore concentrate spillage from trucks and background sources was intermediate to those for the test and control cows.

III.

RECOMMENDATIONS

III. RECOMMENDATIONS

The following recommendations are based on the results of this study:

1. Lead ore concentrate should not be hauled by trucks on public highways or property in open hoppers, or without enough moisture to prevent escape of dust in transit.
2. Dust from stockpiled lead ore concentrate and metallurgical fumes in smelter emissions should be controlled to as small amounts as possible.
3. Tolerances should be determined for lead and cadmium in ambient air and continuous air sampling in lead production areas should be conducted using standard criteria.
4. It should be determined if slag, the end by-product resulting from the smelting process, contains lead which is available to plants and animals.
5. The use of slag in road maintenance should be discouraged because feasible environmental monitoring usually relies on testing soil or other environmental samples, and current analytical methods can not determine with reliability that portion of the total lead that is taken up by plants.
6. Tolerances and/or guidelines should be developed for permissible levels of lead and cadmium in meat and milk products in the United States.

7. A long term surveillance program should be developed to monitor for build up of lead and cadmium in the environment and possible harmful botanical and health effects using soil, cattle hair and other biological samples as markers.
8. Systems to identify sources of lead contamination using lead isotopic ratios of selected food chains involving domestic animals and wildlife in south-eastern Missouri should be developed.
9. Long range plans for uses of National Forests, National Parks and other public lands that involve industrial leasing and development should include specific tolerances and guidelines that will prevent the occurrence of ecological imbalances.

IV.

MATERIALS AND METHODS

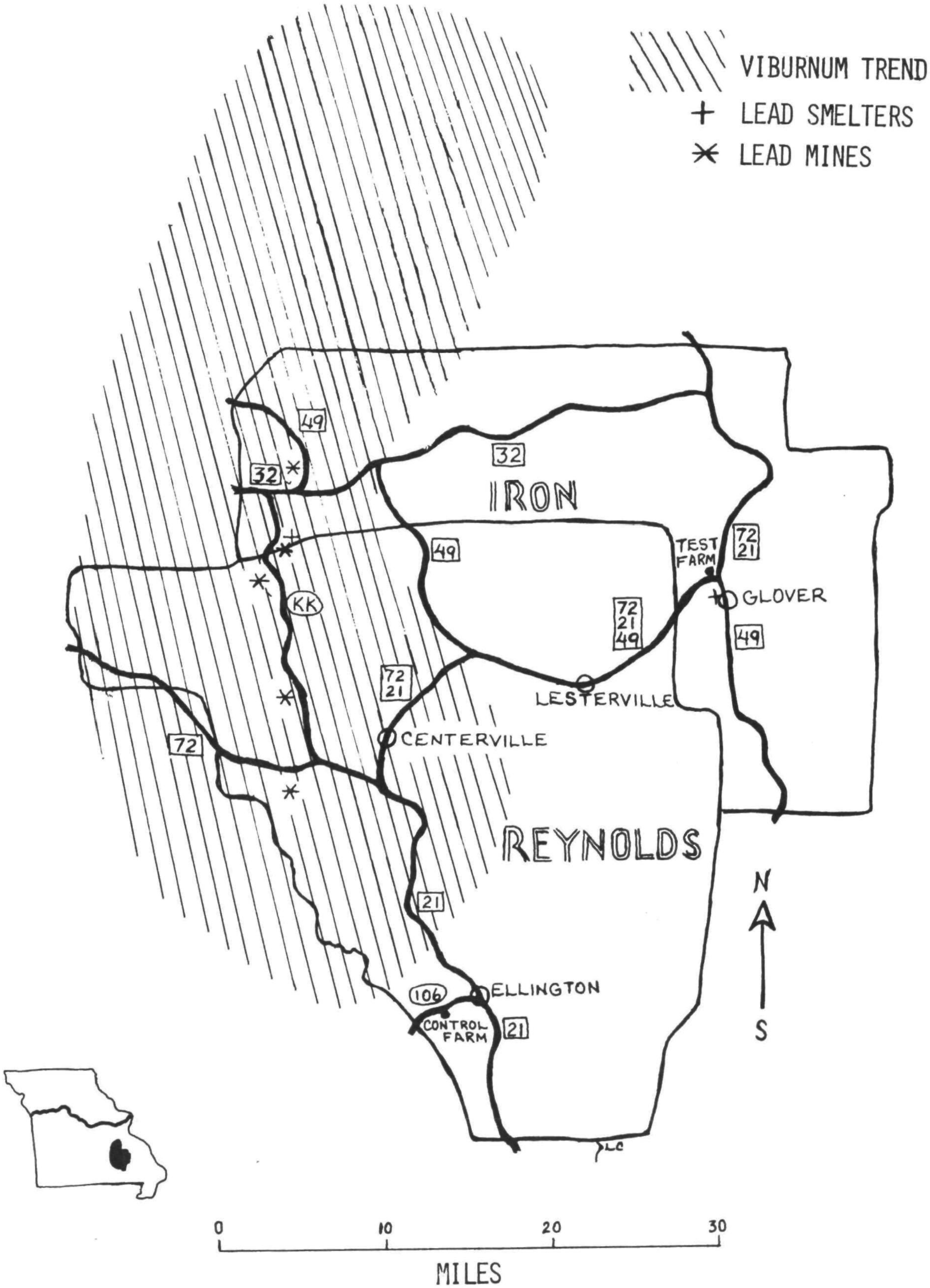
IV. MATERIALS AND METHODS

A. Description of Study Area

The area chosen for study was Iron and Reynolds Counties in southeastern Missouri (Figure 1). Geological exploration of this part of the State in the 1950's and early 1960's indicated that major deposits of lead, zinc, copper and silver were present. Since then extensive industrial development has occurred, and it has become the world's largest lead-producing district. In 1970 the production was 432,576 tons of lead or 74.4% of the entire U.S. lead production.[5] The location of this new mining district has been named the "Viburnum Trend" or "New Lead Belt" to distinguish it from the older lead producing areas that are now inactive.

The development of new mines has been accompanied by the building of two new lead smelters which became operational in 1968. One smelter was built at a mining site in the Clark National Forest,[6] and the other was built on privately owned land at Glover, Missouri. Following milling and concentration, lead ore is hauled by truck via State Highway 72/21 to the smelter at Glover from the mining sites in the Clark National Forest to the west (Figure 1). This part of the highway traverses wooded and agricultural land. The deforested areas are used as pasture for livestock. The chief agricultural products are hay, beef cattle and hogs. Dairy cattle, horses and garden crops are also raised on a more limited basis.

FIGURE 1. A MAP OF IRON AND REYNOLDS COUNTIES IN SOUTHEASTERN MISSOURI SHOWING ROADS, TEST AND CONTROL FARMS, VIBURNUM TREND, LEAD MINES, AND SMELTERS. (MODIFIED FROM HAYES AND SEARIGHT, 1969)



There are several ways in which heavy metals from the lead producing operations enter the environment. The galena (PbS) deposits are approximately 700-1200 feet deep so the metallurgical dusts associated with removal of the ore and milling are limited to the shaft sites. Excess water from the mines and waste water containing tailings are directed to settling and treatment lagoons which have varying efficiencies in removal of heavy metals before discharge into receiving streams in the area.[7] Spillage of lead ore concentrate, which is comprised of approximately 70.0% Pb, 1.5% Zn, 0.5% Cu and 0.2% Cd, from the trucks enroute to the Glover smelter adds to the background levels along the roadsides from automobile emissions and other sources.[8] In the Glover area, especially downwind from the smelter, emissions from the smelter stack and dust from stockpiled ore are sources of contamination. Slag from the smelter, containing approximately 2.5% Pb, is used as road resurfacing material and, also, for road clearing in the winter.

B. Selection of the Test Farm

The selection of the test farm was based upon the availability of cattle for testing and exposure to contamination from production sources of heavy metals. The farm chosen was on the ore trucking highway (72/21) near Glover in Iron County, approximately 800 meters (0.5 mile) north of the smelter stack (Figure 1). During 1970, horses on this farm and a farm on the opposite side of the highway developed signs of lead poisoning and died.

C. Selection of the Control Farm

The control farm was selected from the same geographical region, with as many of the characteristics of the test farm as possible except exposure to lead production sources of contamination. Both the test and control farms have soil of the Clarksville stony loam type, and they have similar topographic features. Results of macroelement, pH and organic matter analyses of soil samples from each farm are presented in Table 1. There was considerable variation between measurements at the beginning and those at the end of the study. This variation, partially due to variation in sample collection and to crop depletion, was approximately as great as the variation between farms. Therefore, the farms were relatively similar in respect to macroelements, pH and organic matter. The depletion of macroelements and organic matter and the decline in pH from the beginning to the end of the study on both farms is understandable in view of the absence of fertilizer applications since 1970, absence of lime applications since 1969 (test farm) and 1966 (control farm), and very little manure application.

Similar grasses were identified (Table 2) and the top growth during 1972 was approximately 1.1 meters on both farms. The amount of rainfall was approximately the same on both farms during the study period (Table 3). The test and control farms were also similar in regard to proximity of pastures to the highway and farming practices. There was a negative

Table 1. History of Fertilizer and Lime Applications and Results of Analyses of Soil Samples from Test and Control Farms

	N	P_2O_5	K	Ca	Mg	$CaCO_3$	pH	Organic matter (percent)
Test Farm								
Last fertilizer application (12-12-12 in 1970)	116	116	116					
Last lime application (red lime in 1969)						8,000		
Soil test, Sep., 1971		58.0	380	3500	600		6.2	5.1
Soil test, Oct., 1972		69.0	210	2200	420		4.9	4.5
Control Farm								
Last fertilizer application (12-12-12 in 1970)	100	100	100					
Last lime application (red lime in 1966)						6,000		
Soil test, Sep., 1971		8.0	95	5100	800		6.3	6.5
Soil test, Oct., 1972		50.0	480	2700	460		5.8	4.1

Table 2. Listing of Grasses in Pastures on Test and Control Farms

Species	Common Name	Test Farm	Control Farm
Poa annua	Blue grass	X	X
Tridens flavus	Purple Top	X	X
Lespedeza virginica	Bush Clover	X	X
Trifolium repens	White Clover	X	
Trifolium pratense	Red Clover	X	

Table 3. Rainfall Recorded on Test and Control Farms during Study Period

Time Period	Inches of Rainfall	
	Test Farm	Control Farm
Total (Oct. 1971 - Sep. 1972)	33.27	37.33
Oct. - Dec. 1971	7.11	9.07
Jan. - Mar. 1972	4.31	3.45
Apr. - Jun. 1972	9.15	12.96
Jul. - Sep. 1972	12.70	11.85

slope away from the roadway: approximately 1.5 meters per 50 on the test farm and approximately 6 meters per 50 on the control farm.

A Highway Commission Survey conducted in 1970-71, indicated that the traffic passing near the test farm was less than that passing a point near the control farm (Table 4). Because of the location of the counters and higher traffic volume near town, the count near the test farm is probably an underestimate and the count near the control farm is probably an overestimate of the volumes that actually pass the test and control farms.

D. Description of the Test and Control Cows

The cows selected for study on the control farm will be described first. The owner-operator of the control farm had a 6 cow dairy herd which provided milk for the family's use and for sale to the local neighbors. The cow that the owner was willing to sell to the project (for slaughter at the end of the study) was a purebred Holstein, 10 years of age. A 10 year old half-sister of this cow, originally from this herd, was purchased from a neighbor and used as a genetically matched comparison cow on the test farm. The youngest of the other cows in the control farm were chosen as the remaining 3 of the required 4 cows to be studied. They were a 3 year old Holstein, a 3 year old Guernsey and a 6 year old Guernsey-milking shorthorn crossbred cow. None of these cows had been moved from the farms so they had never been exposed to lead mining sources of heavy metals.

Table 4. Vehicular Traffic by Season Near Test and Control Farms

	Number of Vehicles*	
Months	Test Farm**	Control Farm+
Oct. - Dec.	76077	136260
Jan. - Mar.	58706	114425
Apr. - Jun.	85107	157202
Jul. - Sep.	96196	177654

*Factored average traffic volume for 1970-1971 from Missouri State Highway Commission data.

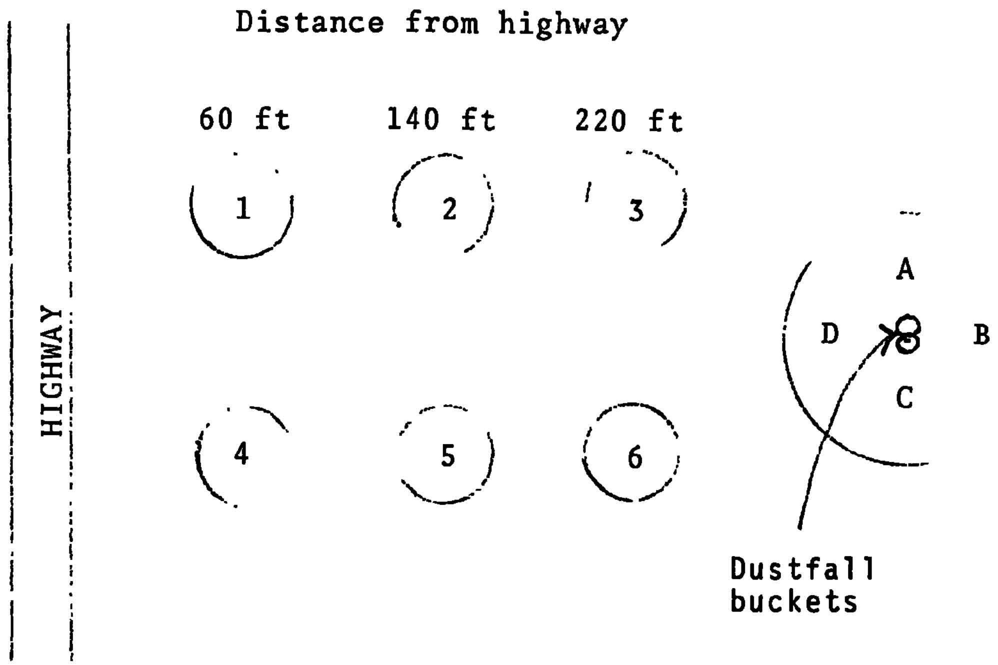
**Counter located on Route 21, .3 miles north of test farm.

+Counter located on Route 106, .3 miles west of Ellington and 3.7 miles east of the control farm.

The test farm was primarily a cow-calf operation with a few pleasure horses for riding. At the initiation of the study, the owner of the farm acquired 2 Hereford-dairy crossbred cows with young calves and a Jersey cow with a young calf. The crossbred cows were 3 years and 5 years of age and the Jersey was approximately 7 years of age. The fourth cow for study on the test farm was the half-sister of the 10 year old Holstein on the control farm. The mean ages were: 6.25 years for the test cows and 5.5 years for the control cows. All of the cows studied on the test farm originally came from locations outside the new lead-belt area, and they were placed on the test farm 1-2 weeks before the first sample collection, October 2, 1971.

E. Sample Collection and Preparation

The main sampling was performed on a seasonal basis: fall (Oct.-Dec.), winter (Jan.-Mar.), spring (Apr.-Jun.), and summer (Jul.-Sep.). This sampling was performed in the same manner on each farm on the same day or on contiguous days. For dustfall, soil, roots and washed and unwashed vegetation tops, 3 distances from the highway were used: 60 ft (18.3 m), 140 ft (42.7 m), and 220 ft (67.1 m). Two rows of 3 sites each were established on each farm as shown in the following site diagram:



1. Dustfall

Settleable particulates were collected in 5.6 liter polyethylene Tupperware™ canisters. These containers were chosen because they are the standard equipment used by the Missouri Air Conservation Commission and other official agencies. The area of the mouth of the container was 0.0284 square meters.

Paired dustfall containers were placed in a holder approximately 1.7 meters above the ground at each of the 6 sampling sites on each farm. Use of both containers was alternated between rows each sampling period to accomplish 50% duplicate sampling. In other words, at each sampling, 9 dustfall containers were collected on each farm (6 from one row and 3 from the other).

Each container was washed with 1% nitric acid and deionized-distilled water 3 times in the field at the time of placement at the site. The dustfall containers were left at each site for approximately 3 months during each sampling period. Ethylene glycol was added to the containers during the winter months to avoid freezing of contents and subsequent cracking of the container.

The contents of each container were emptied into 600 ml Pyrex beakers and, when required, reduced in volume to about 150 ml by boiling. Each container was scrubbed and rinsed with deionized-distilled water to ensure complete removal of contents. Deionized-distilled water was added to make 250 ml total; 25 ml were then transferred to Kjeldahl flasks for wet ashing using a hot acid mixture (5 parts nitric acid : 1 part perchloric acid). The ash was then diluted to 25 ml with 1% nitric acid and stored to await analysis by atomic absorption spectrophotometry (AAS).

2. Air filters

Suspended particles were collected on 2 inch diameter glass fiber filters, Gelman Type E, with efficiency greater than 98% for particles 0.05 microns or larger and a flow rate of approximately 0.025 cubic meter per minute. Collections were made for approximately one month during each of the 4 sampling period at a single site on each farm.

Each pump was calibrated before each new collection, and the air flow during the collection period was calculated. The filters were placed approximately 1.7 meters above the ground with an inverted plastic container mounted over the filter to protect against rain and to avoid collecting large particles that would fall directly on the filter. Clocks were wired into the electrical supply for the air pumps and corrections were made for periods that the electric power was off.

The exposed filter samples were leached using successive amounts of hot acid mixture. Thirty ml of the acid mixture was placed with the filter in a 250 ml beaker with a watch glass cover and boiled for 2 hours. After cooling, the acid was poured off and 30 ml of the acid mixture was added, and the boiling was repeated. The supernatant was again poured off, added to the original supernatant and reduced to ash by heating in Kjeldahl flasks. The ash was then diluted to 25 ml with 1% nitric acid and stored.

3. Soil

Four core samples of the top 15 cm of soil were collected with a soil probe at locations A, B, C and D shown in the site diagram. They were placed in a new polyethylene bag, mixed and then passed through a 12 mesh sieve to remove rocks. At each sampling, one or the other row was alternately duplicated. After drying in an oven at 105° C, a 3 gm portion was refluxed with 20 ml concentrated nitric acid in a 250 ml Pyrex beaker with a watch glass cover. The sample was then passed through a Watman #42 filter and diluted to 100 ml with deionized-distilled water after proper washing.

4. Vegetation-roots

Vegetation roots in 4 locations shown in the site diagram (A, B, C and D) were exposed by a shovel, shaken to remove excess dirt, scrubbed with a polyethylene brush and water, and rinsed with deionized-distilled water. The sample was then placed in tared polyethylene bags and oven dried at 105^{0} C. The dried sample in the bag was then weighed; the weight of the bag was subtracted to obtain the dried sample weight. The sample was then placed in Kjeldahl flasks for wet ashing.

5. Vegetation - tops

Clump-size grass tops were cut 5 cm above the soil at 4 locations (A, B, C and D) shown in the site diagram and composited as one sample in polyethylene canisters containing 200 ml deionized-distilled water. At each sampling, one row or the other was alternately duplicated. The tops were washed in the canisters, transferred to polyethylene bags, oven-dried, weighed as for root samples, and wet ashed. The washings were acidified to 1 N with nitric acid, stored for 1-2 days, and filtered through Watman #42 filter paper. The filtered samples were then analyzed for the specific element. For unwashed vegetation values, the washings and corresponding washed vegetation values were added together.

In addition, green bean, tomatoe and bell pepper samples were collected from home gardens in the vicinity of the test and control farms. They were washed, dried, weighed and wet ashed as for the foliage. Their washings were not analyzed.

6. Cattle hair

Approximately 10 gm of hair was clipped in equal amounts from 4 different areas of the cow's body and then split into duplicate samples in polyethylene bags. Each sample was washed with 1% Snoop* solution, a low trace metal containing soap. The hair was then rinsed twice with deionized-distilled water, air dried, and wet ashed.

7. Cattle blood and milk

Two 5 ml blood samples were collected by jugular venipuncture in heparinized blood vials from each cow. The samples were mixed gently to prevent clotting and stored at 4^{o} C.

Prior to collecting milk samples, the udder of the cow was brushed to remove loose debris and the teats were washed with water. The first milk (approximately 100-200 ml) was discarded. Duplicate milk samples, consisting of milk from each quarter, were collected in 100 ml glass bottles and stored at 4^{o} C.

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*Applied Laboratories, Inc.

The 5 ml samples of blood and milk were added to 30 ml of acid mixture in Kjeldahl flasks for wet ashing. Following cooling the sample was diluted with 1% nitric acid to a final volume of 50 ml.

All of the blood and milk samples were extracted by the following procedure, modified from Pierce and Cholak.[9] One-half ml perchloric acid and 2.0 ml ammonium citrate solution were added to the samples. Ammonium hydroxide or nitric acid was used to adjust the pH to 4.5. After transfer to a 50 ml volumetric flask, 2.0 ml ammonium pyrrolidine dithiocarbamate and 4.0 ml methyl isobutyl ketone, were added to the flask. The organic layer which forms is used for the atomic absorption analysis.

8. Cattle tissues

Five gm specimens of diaphragm muscle, bone, liver and kidney cortex were collected at slaughter of one cow from the test farm and one cow from the control farm. These samples were stored at 4^{o} C and then wet ashed as previously described.

F. Atomic Absorption Spectrophotometry Procedures

The operation of the AAS followed previously described procedures.[10-12] All determinations were made by direct aspiration of the sample solution or the extract into the flame of the atomic absorption unit. Analysis of standards, made according to sample concentration ranges, accompanied each group of samples tested. The concentrations of elements under study were determined in blood vial, ethylene glycol and glass fiber filter blanks (Table 5). All of the measurements were low except for the zinc content in the glass fiber filters. The mean filter blank zinc concentration (581 ug/gm) was subtracted from the sample measurements to obtain corrected values which were used in reporting the results. All values are expressed on a dry weight basis except for blood, milk, muscle, liver, kidney cortex and bone which are on a fresh weight basis. The minimum detectable limits for specific types of samples and elements are presented in Table 6.

G. Statistical Procedures

All samples were coded prior to submitting them to the laboratory so that the laboratory personnel had no knowledge of the study design considerations in the sampling. The identifying information for each sample and the concentration values were coded and placed on punch cards for statistical analysis. All data were normalized by log transformation to satisfy necessary assumptions for analysis of variance (ANOVA). ANOVA was performed to determine the effect of farm location, season and sampling site upon the concentrations of Cd, Cu, Pb and Zn in the various components of the food chain under study.

The ANOVA model for dustfall, soil and vegetation was:

$$W_{ijkl} = u + T_i + P_j + TP_{ij} + D_k + DT_{ik} + DP_{jk} + DTP_{ijk} + M_{ijkl}$$

where:

u represents some overall element measurement in ug/gm

T_i represents the effect of the i^{th} time period on the measurement: i = 1, 4

Table 5. Concentrations of Cd, Cu, Pb and Zn in Blood Vials, Ethylene Glycol and Glass Fiber Filters

Blanks	Element (ug/gm)			
	Cd	Cu	Pb	Zn
Blood vials	<.01	<.01	<.01	1.04
Ethylene glycol	<.10	<.10	<.08	<.10
Glass fiber filters	<.25	.65	<2.50	581

Table 6. MINIMUM DETECTABLE LIMITS BY TYPE OF SAMPLE AND ELEMENT

Code No.	Type of Sample	Element	Minimum Detectable Limit
1	Air Sample Filter	Cd	0.50 ug
4	Dustfall	Cd	2.5 ug
5	Soil	Cd	0.35 ug/gm
6	Roots (washed)	Cd	0.50 ug/gm
		Pb	5.0 ug/gm
7	Tops (washed)	Cd	0.50 ug/gm
		Pb	5.0 ug/gm
8	Tops (unwashed)	Cd	1.5 ug/gm*
		Cu	1.0 ug/gm*
		Pb	15.0 ug/gm*
9	Blood	Cd	0.2 ug/100 ml
		Pb	2.0 ug/100 ml
10	Milk	Cd	0.2 ug/100 ml
		Cu	7.0 ug/100 ml
		Pb	2.0 ug/100 ml
11	Hair	Cd	0.01 ug/gm
12	Muscle	Cd	0.10 ug/gm
13	Bone	Cd	0.05 ug/gm
17	Water	Cd	0.01 mg/l
		Cu	0.01 mg/l
		Pb	0.005 mg/l
		Zn	0.01 mg/l
18	Grain Feed	Cd	0.50 ug/gm
		Pb	5.0 ug/gm
19	Hay or Silage	Cd	0.50 ug/gm
		Pb	5.0 ug/gm
21	Home-grown Vegetables	Cd	0.50 ug/gm
		Pb	5.0 ug/gm

*If both washed tops and washings were below minimum detectable limits.

P_j represents the effect of the j^{th} farm on the measurement:

$j = 1, 2$

TP_{ij} represents the effect on the ij^{th} time period-farm combination on the measurement

D_k represents the effect of k^{th} distance (site) from the highway: $k = 1, 3$

DT_{ik} represents the effect of the ik^{th} time period-distance combination on the measurement

DP_{jk} represents the effect of the jk^{th} farm-distance combination on the measurement

DTP_{ijk} represents the effect of the ijk^{th} time period-farm-distance combination on the measurement

M_{ijkl} represents the error term: $l = 1, 3$

W_{ijkl} represents ug/gm of a specific element in dustfall, soil, roots, unwashed vegetation tops and washed vegetation tops

The conventional 5% level (p=0.05) significance was used in interpreting the results. The p values are expressed to the fourth decimal, although an individual value may have little meaning beyond the second decimal because of the approximations involved in the statistical analysis. Because of the large number of comparisons and statistical tests performed, it should also be recognized that even if no effects were present, some differences may be statistically significant due to chance alone.

The ANOVA model for hair, blood, milk, muscle and bone was:

$$X_{ijkl} = u + T_i + P_j + TP_{ij} + C_{k(j)} + CT_{ik(j)} + M_{ik(j)l}$$

where:

u represents some overall element measurement in ug/gm

T_i represents the effect of the i^{th} time period on the measurement: $i = 1, 4$

P_j represents the effect of the j^{th} farm on the measurement: $j = 1, 2$

TP_{ij} represents the effect of the ij^{th} time period-farm combination on the measurement

$C_{k(j)}$ represents the effect of the k^{th} cow on the j^{th} farm on the measurement $k = 1, 4$

$CT_{ik(j)}$ represents the effect of the ik^{th} time period-cow combination of the j^{th} farm on the measurement

$M_{ik(j)l}$ represents the error term: $l = 1, 2$

X_{ijkl} represents ug/gm of a specific element in blood, hair milk, muscle or bone.

For the tabular presentations of mean concentrations of a specific element, individual values below the lower detectable limit for that element and type of sample (Table 6) were excluded, except for unwashed samples of vegetation tops. In the analysis of variance calculations, the maximum values were used for samples that contained a smaller concentration than the lower detectable limit. For example, the cadmium value of 0.35 ug/gm was used for soil samples containing less than the lower detectable limit. This has the result of avoiding assumptions of the exact values for those lower than the detection limit. The control farm samples were more often below lower detection limits than test farm samples; therefore, some comparisons between test and control farm samples are conservative.

V.

RESULTS AND DISCUSSION

V. RESULTS AND DISCUSSION

A summary of variables with significant effects on heavy metal concentration in the various food chain components studied is presented in Table 7. Results of analyses of each component will first be reported and then inter-relationships will be considered.

A. Dustfall

The dustfall concentrations of all 4 elements (Cd, Cu, Pb and Zn) were significantly different on the test and control farms (Tables 8-11). The test farm to control farm ratios for each element were: cadmium 11.9, copper 5.9, lead 12.9, and zinc 6.8. Cadmium and zinc concentrations in dustfall varied significantly between seasons. The winter season dustfall samples from the test farm had the highest levels of Cd, Cu and Pb; Zn was highest in the summer. The results indicated that distance from the highway was a significant effect in the Cu and Zn analyses. Copper was in highest concentration at the 140 ft site on the control farm during all seasons, while on the test farm there was a gradient of copper levels from a high at the 60 ft site to a low at the 220 ft site in all seasons except fall (Appendix Tables 1-8). An extremely high lead dustfall measurement (170.3140 $mg/m^2/mo$) was obtained at the 60 ft site on the test farm in the winter (Appendix Table 3). There was a significant interaction of farm and season in the cadmium analysis.

B. Soil

The soils of the test farm and of the control farm significantly differed in their concentrations of each of the elements studied: Cd, Cu, Pb and Zn (Tables 12-15). In each comparison, the concentration was greater on the test farm than on the control farm. This was found for Zn even though the fall Zn concentration on the control farm was much higher than that on the test farm. From these fall data (Appendix Tables 1-2) it was apparent that an aberrant source of Zn was present and further examination of the data revealed that a 60 ft site near a galvanized fence yielded high Zn levels in soil, as well as in roots and vegetation tops. All subsequent samples were collected at distances greater than 6 feet from the fence.

There was significant seasonal variability in the concentrations of Cd, Cu and Pb in soil. The soil lead concentrations increased over 2 fold during the study period on the test farm.

The distance from the highway also had a significant effect on the soil concentration of all 4 elements. For each element, there was a gradient from high values at the 60 ft site to lower values at the 220 ft site, except for unexplained high levels of copper at the 140 ft site on the control farm in all 4 seasonal samples (Appendix Tables 2, 4, 6 and 8). These differences in copper levels in soil corresponded to the copper levels in dustfall and indicated that copper was a highway contaminant on the test farm but not on the control farm.

There were significant farm-season interactions for Cd, Pb and Zn measurements which may reflect in part the seasonally determined meterological effects on the distribution of smelter stack emissions at the test farm. Farm and site interactions were significant in Cd, Cu and Pb analyses. This was expected in view of the multiple sources of highway contamination on the test farm compared with only vehicular exhaust emissions as a main source on the control farm.

C. Vegetation - roots

The Cd, Cu, Pb and Zn concentrations in roots all differed significantly on the test and control farms (Tables 16-19). There was also a significant effect of season on the concentrations of all 4 elements. Only Pb and Zn were significantly affected by distance from the highway. An extremely high mean zinc concentration (259.33 ug/gm) in 3 root samples from the 60 ft site at the control farm in the fall, was attributed to the galvanized fence (Appendix Table 2). While the cadmium in soil ratio on test and control farms was approximately 1:1, the comparable cadmium in root ratio was approximately 3:1. The lead in soil ratio was similarly smaller than the lead in root ratio on test and control farms (5:1 vs. 12:1).

D. Vegetation - tops

The differences between the test and control farms' concentrations of all 4 elements, Cd, Cu, Pb and Zn, in unwashed vegetation tops were significant (Tables 20-23). The largest differences were for Cd and Pb. Lead in unwashed tops on the test farm was approximately 19.6 times higher than on the control farm and cadmium was approximately 3.6 times higher than on the control farm. All 4 elements also differed significantly from season to season. The zinc variability in unwashed tops is again probably due to sample collection near the galvanized fence, as the extremely high mean value of 337.37 ug/gm was obtained in the fall on the control farm (Appendix Table 2). On the test farm, winter and spring were consistently higher than summer and fall. High concentrations of heavy metals in foliage during winter periods when plants are dormant have been observed in several other studies.[13-14] The spring sample was collected on April 1 when the grass was growing again, but before the extensive growth and grazing which followed. In general, the pasture was grazed so that most grasses did not grow higher than 60 cm, except where the cattle were restricted and the grass grew to 1.1 meters.

Only zinc concentrations in unwashed tops were significantly different on test and control farms. An inspection of the data again implicated the samples, collected near the fence, that contributed the high values. Farm-season interaction was significant for all 4 elements. The higher contamination from multiple sources at the test farm, linked with the seasonal effect of dormant plants having higher concentrations than rapidly growing plants, could explain this interaction.

The levels of all 4 elements in washed tops varied significantly between test and control farms (Tables 24-27). All elements except zinc were found in much higher amounts on the test farm than on the control farm. The high zinc levels along the fence on the control farm again appeared to affect the levels in the washed vegetation. Also, the significant farm-site interaction reflects this unique exposure on the control farm. The elemental concentrations in washed tops were uniformly affected by seasons. Most of this effect was present on the test farm where all 4 elements were highest in tops during the winter and spring test periods. The site variable was not significant for any element tested in washed tops. The only significant interaction, farm and season, could be explained by the unique exposure to smelter and trucking contamination by Cd, Cu and Pb on the test farm and to galvanized fence contamination by Zn on the control farm.

E. Cattle Hair

The concentrations of Cd and Pb in the washed hair of the 4 cows on the test farm were significantly different than the concentrations in hair of the 4 control cows (Tables 28 and 30). At the summer sampling the Cd concentration in the project owned test cow's hair was approximately 10 times higher than that of the project owned control cow's hair, and the Pb concentration in the test cow's hair was approximately 115 times higher than that of the control cow's hair (Appendix Tables 7-8).

The hair concentrations of three elements, Cd, Pb and Zn, were significantly affected by season (Tables 28, 30-31). Only Cu and Zn hair concentrations varied significantly among the cows on each farm (Tables 29 and 31).

Even though the hair was washed, apparently it is possible that cadmium and lead adsorbed by hair remains in hair after the washing process. Nishujama reported that cadmium and lead adsorbed on human and mouse hair was incompletely removed by different treatments.[15] The most complete removal of cadmium was by using a sufficiently strong solution of an acid; however, the different treatments were not effective for separate analysis of exogenous and endogenous cadmium in hair.

Therefore, analysis of hair concentrations of cadmium and lead may not truly represent body accumulation, depending upon the amount of airborne exposure. In the present study airborne exposure was negligible on the control farm and uniformly high on the test farm. The higher hair cadmium and lead concentrations on the test farm than those on the control farm, therefore, may reflect both the increased lead and cadmium assimilation by the cattle and the adsorption of airborne cadmium and lead on the hair. In any event, the high hair concentrations of both elements truly reflected high airborne concentrations on the test farm. That hair lead concentrations were higher than the concentrations of any of the other biological samples tested (Figure 4), supports the use of cattle hair as a sensitive indicator of airborne lead contamination.

F. Cattle Blood

The cows' blood Pb and Cu concentrations were both significantly different on the test and control farms (Tables

33-34). The mean of the blood lead concentrations of the test cows was approximately 4 times greater than the corresponding mean value for the control cows. The difference between blood copper concentrations of test and control cows was in the opposite direction, i.e. the control cows were higher than the test cows.

Blood concentrations of Cd, Cu and Pb for test and control cows were significantly affected by season (Tables 32-34). The only significant variability detected among cows, within farm, was detected for blood zinc concentrations (Table 35).

The highest concentration of blood lead was a mean of 87 ug/100 ml for 2 duplicate analyses of a spring blood sample collected from the youngest (3 year old) cow on the test farm (Appendix Table 5). The relationship between blood lead and hair lead for the test cows is shown in Figure 2.

G. Milk from Cows

Lead was the only element studied that was significantly affected by season and the only one present in significantly different concentrations in milk from test and control cows (Tables 36-39). The lead concentration was over 5 times higher in milk from the project owned test cow than milk from the project owned control cow. The highest concentration of milk Pb was a mean of 35 ug/100 ml for 2 duplicate analyses performed on a sample collected April 1 (spring) from the 7 year old cow on the test farm. As found for hair and blood, the concentrations of cadmium varied significantly among cows on each farm.

There was significant interaction between farm and season variables in the lead analysis. The winter and spring milk lead concentrations were over 5 times the fall and summer concentrations on the test farm only. The relationship between milk Pb and blood Pb for the test cows is shown in Figure 3.

Table 7. Summary of Farm Location (F), Season (S), Gradient Distance from Highway (D) and Cow Within Farm (C) Variables With Significant* Effect on Heavy Metal Concentrations in Each Type of Environmental Sample

Type of Sample	Cadmium	Copper	Lead	Zinc
Dustfall	F, S	F, D	F	F, S, D
Soil	F, S, D	F, S, D	F, S, D	F, D
Roots	F, S	F, S	F, S, D	F, S, D
Unwashed tops	F, S	F, S	F, S	F, S, D
Washed tops	F, S	F, S	F, S	F, S
Hair	F, S	C	F, S	S, C
Blood	S	F, S	F, S	C
Milk		C	F, S	

*Statistically significant at the 5% level

Table 8. Summary of Data and Analysis of Variance for Cadmium in Dustfall

Season	Test Farm: No. of Samples**	Test Farm: Mean ($mg/m^2/mo$)	Control Farm: No. of Samples**	Control Farm: Mean ($mg/m^2/mo$)
Fall	8	0.7484	7	0.2427
Winter	9	1.6617	1	0.0386
Spring	8+	0.5439	9	0.0528
Summer	9	1.3913	2	0.0322

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	452.619	107.981	.0001
Season	3	41.481	3.299	.0272
Site	2	1.207	0.144	.8665
Farm*Season	2	60.868	7.261	.0021
Farm*Site	2	1.900	0.227	.8005
Season*Site	6	24.044	0.956	.5346
Farm*Season*Site	4	10.728	0.640	.6395
Residual (error)	50	209.583		

**Samples below lower detectable limit (2.5 ug) excluded; analysis of variance based on maximum values for sample tests that were below lower detectable limit.

+One sample lost in laboratory preparation.

Table 9. Summary of Data and Analysis of Variance for Copper in Dustfall

	Test Farm		Control Farm	
Season	No. of Samples	Mean ($mg/m^2/mo$)	No. of Samples	Mean ($mg/m^2/mo$)
Fall	9	2.2155	9	0.7602
Winter	9	2.7543	9	0.1983
Spring	8**	2.2300	9	0.3332
Summer	9	1.9682	9	0.2515

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	75.003	216.688	.0001
Season	3	0.817	0.787	.5097
Site	2	2.408	3.479	.0374
Farm*Season	2	0.212	0.306	.7419
Farm*Site	2	0.960	1.387	.2582
Season*Site	6	1.834	0.883	.5152
Farm*Season*Site	4	0.625	0.452	.7730
Residual (error)	50	17.307		

**One sample lost in laboratory preparation.

Table 10. Summary of Data and Analysis of Variance for Lead in Dustfall

Season	Test Farm No. of Samples	Test Farm Mean (mg/m^2/mo)	Control Farm No. of Samples	Control Farm Mean (mg/m^2/mo)
Fall	9	97.6796	9	25.9181
Winter	9	141.4106	9	2.1435
Spring	8**	86.0278	9	2.8856
Summer	9	96.4958	9	1.7206

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	218.296	161.944	.0001
Season	3	7.701	1.904	.1396
Site	2	0.668	0.248	.7844
Farm*Season	2	0.538	0.200	.8214
Farm*Site	2	1.026	0.381	.6908
Season*Site	6	5.558	0.687	.6628
Farm*Season*Site	4	0.725	0.135	.9661
Residual (error)	50	67.398		

**One sample lost in laboratory preparation.

Table 11. Summary of Data and Analysis of Variance for Zinc in Dustfall

Season	Test Farm No. of Samples	Test Farm Mean (mg/m^2/mo)	Control Farm No. of Samples	Control Farm Mean (mg/m^2/mo)
Fall	9	1.4479	9	1.8170
Winter	9	14.4001	9	1.2800
Spring	8**	12.3750	9	1.8486
Summer	9	16.2486	9	1.5559

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	47.299	444.499	.0001
Season	3	36.504	114.349	.0001
Site	2	0.975	4.582	.0147
Farm*Season	2	0.497	2.335	.1053
Farm*Site	2	0.240	1.130	.3316
Season*Site	6	0.622	0.974	.5464
Farm*Season*Site	4	0.100	0.234	.9164
Residual (error)	50	5.321		

**One sample lost in laboratory preparation.

Table 12. Summary of Data and Analysis of Variance for Cadmium in Soil

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples**	Control Farm Mean (ug/gm)
Fall	9	.56	9	.40
Winter	9	.91	9	.60
Spring	9	.70	1	.57
Summer	9	.83	1	.37

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	5.399	342.001	.0001
Season	3	2.039	43.055	.0001
Site	2	0.126	4.006	.0240
Farm*Season	3	0.745	15.721	.0001
Farm*Site	2	0.118	3.736	.0302
Season*Site	6	0.105	1.109	.3711
Farm*Season*Site	6	0.093	0.982	.5511
Residual (error)	48	0.758		

**Samples below lower detectable limit (0.35 ug/gm) excluded; analysis of variance based on maximum values (0.35 ug/gm) for sample tests that were below lower detectable limit.

Table 13. Summary of Data and Analysis of Variance for Copper in Soil

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	9.86	9	5.71
Winter	9	12.01	9	7.58
Spring	9	12.76	9	7.82
Summer	9	12.26	9	6.50

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	5.039	273.746	.0001
Season	3	0.837	15.160	.0001
Site	2	0.334	9.071	.0007
Farm*Season	3	0.088	1.599	.2008
Farm*Site	2	0.376	10.204	.0004
Season*Site	6	0.034	0.310	.9282
Farm*Season*Site	6	0.171	1.545	.1836
Residual (error)	48	0.884		

Table 14. Summary of Data and Analysis of Variance for Lead in Soil

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	52.00	9	13.00
Winter	9	77.83	9	15.13
Spring	9	88.62	9	18.87
Summer	9	128.28	9	15.93

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	45.205	836.47	.0001
Season	3	3.743	23.09	.0001
Site	2	1.291	11.95	.0002
Farm*Season	3	1.551	9.57	.0001
Farm*Site	2	0.681	6.300	.0040
Season*Site	6	0.337	1.040	.4120
Farm*Season*Site	6	0.435	1.342	.2570
Residual (error)	48	55.426		

Table 15. Summary of Data and Analysis of Variance for Zinc in Soil

Samples	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	24.11	9	30.56
Winter	9	30.24	9	22.16
Spring	9	33.16	9	23.89
Summer	9	33.24	9	15.69

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	1.727	27.153	.0001
Season	3	0.447	2.344	.0836
Site	2	1.319	10.368	.0004
Farm*Season	3	1.605	8.410	.0003
Farm*Site	2	0.221	1.740	.1848
Season*Site	6	0.299	0.784	.5884
Farm*Season*Site	6	0.961	2.518	.0333
Residual (error)	48	3.053		

Table 16. Summary of Data and Analysis of Variance for Cadmium in Roots

	Test Farm		Control Farm	
Season	No. of Samples	Mean (ug/gm)	No. of Samples**	Mean (ug/gm)
Fall	9	1.54	8	.92
Winter	9	3.13	9	.65
Spring	9	2.87	8	.69
Summer	9	1.60	8	.72

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	20.742	198.650	.0001
Season	3	1.301	4.155	.0108
Site	2	0.056	0.267	.7699
Farm*Season	3	3.224	10.293	.0001
Farm*Site	2	0.221	1.059	.3559
Season*Site	6	0.859	1.371	.2447
Farm*Season*Site	6	0.785	1.254	.2958
Residual (error)	48	5.012		

**Sample below lower detectable limit (0.50 ug/gm) excluded; analysis of variance based on maximum values (0.50 ug/gm) for sample tests that were below lower detectable limit.

Table 17. Summary of Data and Analysis of Variance for Copper in Roots

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	21.89	9	11.32
Winter	9	33.88	9	20.69
Spring	9	24.28	9	15.71
Summer	9	17.49	9	11.62

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	4.325	84.378	.0001
Season	3	4.197	27.291	.0001
Site	2	0.057	0.559	.5808
Farm*Season	3	0.163	1.063	.3745
Farm*Site	2	0.387	3.779	.0291
Season*Site	6	0.105	0.340	.9117
Farm*Season*Site	6	0.580	1.886	.1021
Residual (error)	48	2.460		

Table 18. Summary of Data and Analysis of Variance for Lead in Roots

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples**	Control Farm Mean (ug/gm)
Fall	9	208.22	9	12.73
Winter	9	238.67	8	8.74
Spring	9	309.00	9	22.33
Summer	9	132.11	8	10.53

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	139.849	606.494	.0001
Season	3	6.091	8.805	.0002
Site	2	1.594	3.456	.0385
Farm*Season	3	1.886	2.726	.0534
Farm*Site	2	1.632	3.539	.0358
Season*Site	6	1.628	1.177	.3341
Farm*Season*Site	6	1.877	1.356	.2507
Residual (error)	48	11.068		

**Sample below lower detectable limit (5.0 ug/gm) excluded; analysis of variance based on maximum values (5.0 ug/gm) for sample tests that were below lower detectable limit.

Table 19. Summary of Data and Analysis of Variance for Zinc in Roots

Season	Test Farm		Control Farm	
	No. of Samples	Mean (ug/gm)	No. of Samples	Mean (ug/gm)
Fall	9	79.89	9	124.56
Winter	9	100.78	9	45.56
Spring	9	84.44	9	38.17
Summer	9	57.89	9	54.61

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	2.583	15.332	.0005
Season	3	1.940	3.839	.0151
Site	2	4.318	12.817	.0001
Farm*Season	3	2.518	4.983	.0046
Farm*Site	2	0.310	0.921	.5924
Season*Site	6	2.504	2.478	.0357
Farm*Season*Site	6	1.795	1.776	.1236
Residual (error)	48	8.085		

Table 20. Summary of Data and Analysis of Variance for Cadmium in Vegetation (unwashed)

Season	Test Farm No. of Samples**	Test Farm Mean (ug/gm)	Control Farm No. of Samples**	Control Farm Mean (ug/gm)
Fall	9	3.60	9	1.74
Winter	9	8.41	9	1.55
Spring	9	8.72	9	1.50
Summer	8+	2.03	9	1.52

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	21.238	311.081	.0001
Season	3	5.816	28.396	.0001
Site	2	0.121	0.885	.5777
Farm*Season	3	6.494	31.708	.0001
Farm*Site	2	0.091	0.664	.5239
Season*Site	6	0.488	1.192	.3265
Farm*Season*Site	6	0.303	0.739	.6227
Residual (error)	47	3.209		

**For calculation of means and analysis of variance, minimum detectable limit values (0.50 ug/gm for washed vegetation; 1.00 ug/gm for washings) were used for measurements below these limits.

+One sample lost.

Table 21. Summary of Data and Analysis of Variance for Copper in Vegetation (unwashed)

Season	Test Farm		Control Farm	
	No. of Samples**	Mean (ug/gm)	No. of Samples**	Mean (ug/gm)
Fall	9	12.63	9	11.85
Winter	9	19.67	9	9.42
Spring	9	17.84	9	7.08
Summer	8+	6.71	9	5.78

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	4.116	75.783	.0001
Season	3	5.684	34.886	.0001
Site	2	0.123	1.129	.3326
Farm*Season	3	2.154	13.221	.0001
Farm*Site	2	0.316	2.906	.0630
Season*Site	6	0.116	0.357	.9021
Farm*Season*Site	6	0.215	0.659	.6845
Residual (error)	47	2.553		

**For calculation of means and analysis of variance the minimum detectable limit value for washings (1.00 ug/gm) was used for sample measurements below this limit.

+One sample lost.

Table 22. Summary of Data and Analysis of Variance for Lead in Vegetation (unwashed)

Season	Test Farm: No. of Samples**	Test Farm: Mean (ug/gm)	Control Farm: No. of Samples**	Control Farm: Mean (ug/gm)
Fall	9	326.00	9	25.74
Winter	9	979.44	9	41.13
Spring	9	823.00	9	37.64
Summer	8+	118.15	9	15.44

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	141.262	726.288	.0001
Season	3	23.928	41.009	.0001
Site	2	0.645	1.657	.2000
Farm*Season	3	4.336	7.431	.0006
Farm*Site	2	0.235	0.604	.5558
Season*Site	6	0.697	0.597	.7330
Farm*Season*Site	6	0.454	0.389	.8826
Residual (error)	47	9.141		

**For calculation of means and analysis of variance, minimum detectable limit values (5.00 ug/gm for washed vegetation; 1.00 ug/gm for washings) were used for sample measurement below these limits.

+One sample lost.

Table 23. Summary of Data and Analysis of Variance for Zinc in Vegetation (unwashed)

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	60.20	9	157.62
Winter	9	82.18	9	78.40
Spring	9	102.88	9	32.39
Summer	8**	36.11	9	31.16

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	1.107	5.416	.0229
Season	3	7.362	12.007	.0001
Site	2	2.442	5.975	.0051
Farm*Season	3	6.272	10.230	.0001
Farm*Site	2	0.593	1.451	.2435
Season*Site	6	0.831	0.678	.6702
Farm*Season*Site	6	1.134	0.924	.5127
Residual (error)	47	9.605		

**One sample lost.

Table 24. Summary of Data and Analysis of Variance for Cadmium in Washed Vegetation

Season	Test Farm: No. of Samples**	Test Farm: Mean (ug/gm)	Control Farm: No. of Samples**	Control Farm: Mean (ug/gm)
Fall	9	2.53	8	.83
Winter	9	6.98	2	.73
Spring	9	7.29	0	--
Summer	7	1.11	1	.70

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	53.420	479.563	.0001
Season	3	10.450	31.271	.0001
Site	2	0.075	0.336	.7209
Farm*Season	3	13.122	39.266	.0001
Farm*Site	2	0.220	0.987	.6179
Season*Site	6	0.476	0.712	.6438
Farm*Season*Site	6	0.682	1.020	.4248
Residual (error)	47	5.236		

**Samples below lower detectable limit (0.50 ug/gm) excluded; analysis of variance based on maximum values (0.50 ug/gm) for sample tests that were below lower detectable limit.

+One value missing.

Table 25. Summary of Data and Analysis of Variance for Copper in Washed Vegetation

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	11.13	9	8.72
Winter	9	14.63	9	6.24
Spring	9	16.11	9	6.06
Summer	8**	5.34	9	4.64

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	5.377	119.268	.0001
Season	3	5.478	40.504	.0001
Site	2	0.192	2.131	.1282
Farm*Season	3	2.463	18.208	.0001
Farm*Site	2	0.112	1.243	.2976
Season*Site	6	0.076	0.282	.9421
Farm*Season*Site	6	0.256	0.948	.5286
Residual (error)	47	2.119		

**One value missing.

Table 26. Summary of Data and Analysis of Variance for Lead in Washed Vegetation

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples**	Control Farm Mean (ug/gm)
Fall	9	267.56	9	13.19
Winter	9	771.67	9	19.36
Spring	9	711.67	9	24.50
Summer	8+	87.54	5	5.80

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	187.765	985.125	.0001
Season	3	33.455	58.509	.0001
Site	2	0.378	0.991	.6194
Farm*Season	3	2.749	4.808	.0056
Farm*Site	2	0.004	0.011	.9902
Season*Site	6	0.626	0.547	.7712
Farm*Season*Site	6	0.499	0.436	.8516
Residual (error)	47	8.958		

**Samples below lower detectable limit (5.0 ug/gm) excluded; analysis of variance based on maximum values (5.0 ug/gm) for sample tests that were below lower detectable limit.

+One value missing.

Table 27. Summary of Data and Analysis of Variance for Zinc in Washed Vegetation

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	9	55.11	9	141.33
Winter	9	61.70	9	48.10
Spring	9	85.67	9	26.56
Summer	8**	29.36	9	26.51

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	1.156	5.358	.0236
Season	3	7.264	11.218	.0001
Site	2	1.247	2.889	.0640
Farm*Season	3	6.012	9.285	.0002
Farm*Site	2	1.270	2.943	.0609
Season*Site	6	1.545	1.193	.3260
Farm*Season*Site	6	1.611	1.244	.3010
Residual (error)	47	10.144		

**One value missing.

Table 28. Summary of Data and Analysis of Variance for Cadmium in Hair

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	8	1.29	8	.06
Winter	8	1.74	8	.13
Spring	8	2.80	8	.05
Summer	8	0.67	7**	.04

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	163.190	423.630	0.0000
Season	3	11.334	9.807	0.0005
Cow within farm	6	1.578	0.683	0.6658
Farm*Season	3	3.211	2.779	0.0710
Season*Cow within farm	18	6.934		
Duplicate analysis error	32	3.003		

**One sample below lower detectable limit excluded; analysis of variance based on maximum value (i.e. 0.01 ug/gm) for the sample test that was below lower detectable limit.

Table 29. Summary of Data and Analysis of Variance for Copper in Hair

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	8	8.26	8	7.25
Winter	8	7.76	8	7.84
Spring	8	6.94	8	6.81
Summer	8	7.99	8	7.41

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.047	2.226	0.1530
Season	3	0.179	2.802	0.0694
Cow within farm	6	0.536	4.196	0.0082
Farm*Season	3	0.041	0.642	0.5979
Season*Cow within farm	18	0.383		
Duplicate analysis error	32	0.356		

Table 30. Summary of Data and Analysis of Variance for Lead in Hair

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	8	94.13	8	2.19
Winter	8	87.50	8	3.92
Spring	8	96.50	8	2.13
Summer	8	66.00	8	0.88

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	233.536	782.272	0.0000
Season	3	7.414	8.278	0.0011
Cow within farm	6	4.317	2.410	0.0691
Farm*Season	3	2.790	3.115	0.0521
Season*Cow within farm	18	5.374		
Duplicate analysis error	32	6.272		

Table 31. Summary of Data and Analysis of Variance for Zinc in Hair

Season	Test Farm No. of Samples	Test Farm Mean (ug/gm)	Control Farm No. of Samples	Control Farm Mean (ug/gm)
Fall	8	104.50	8	93.75
Winter	8	134.88	8	115.88
Spring	8	130.50	8	101.88
Summer	8	93.30	8	82.59

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.096	3.514	0.0772
Season	3	1.694	20.619	0.0000
Cow within farm	6	0.641	3.901	0.0114
Farm*Season	3	0.021	0.252	0.8588
Season*Cow within farm	18	0.493		
Duplicate analysis error	32	2.019		

Table 32. Summary of Data and Analysis of Variance for Cadmium in Blood

Season	Test Farm		Control Farm	
	No. of Samples**	Mean (ug/100 ml)	No. of Samples**	Mean (ug/100 ml)
Fall	8	1.74	8	2.16
Winter	6	0.38	4	0.38
Spring	6	0.60	6	0.80
Summer	7	0.76	8	0.36

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.249	0.437	0.5167
Season	3	29.366	17.195	0.0000
Cow within farm	6	2.537	0.743	0.6226
Farm*Season	3	0.678	0.397	0.7568
Season*Cow within farm	18	10.247		
Duplicate analysis error	32	6.270		

**Samples below lower detectable limit excluded; analysis of variance based on maximum values (i.e. 0.02 ug/100 ml) for sample tests that were below detectable limits.

Table 33. Summary of Data and Analysis of Variance for Copper in Blood

Season	Test Farm No. of Samples	Test Farm Mean (ug/100 ml)	Control Farm No. of Samples	Control Farm Mean (ug/100 ml)
Fall	8	85.88	8	97.25
Winter	8	97.13	8	111.00
Spring	8	63.48	8	109.75
Summer	8	76.13	8	98.13

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	1.117	42.966	0.0000
Season	3	0.448	5.748	0.0061
Cow within farm	6	0.306	1.960	0.1254
Farm*Season	3	0.455	5.839	0.0057
Season*Cow within farm	18	0.468		
Duplicate analysis error	32	0.241		

Table 34. Summary of Data and Analysis of Variance for Lead in Blood

Season	Test Farm No. of Samples	Test Farm Mean (ug/100 ml)	Control Farm No. of Samples	Control Farm Mean (ug/100 ml)
Fall	8	34.00	8	18.88
Winter	8	46.50	8	8.75
Spring	8	58.75	8	10.38
Summer	8	28.13	8	6.63

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	30.168	233.265	0.0000
Season	3	4.071	10.497	0.0003
Cow within farm	6	0.655	0.845	0.5523
Farm*Season	3	3.438	8.864	0.0008
Season*Cow within farm	18	2.327		
Duplicate analysis error	32	1.399		

Table 35. Summary of Data and Analysis of Variance for Zinc in Blood

	Test Farm		Control Farm	
Season	No. of Samples	Mean (ug/100 ml)	No. of Samnles	Mean (ug/ 100 ml)
Fall	8	389.75	8	495.25
Winter	8	391.50	8	376.88
Spring	8	425.25	8	379.13
Summer	8	402.50	8	372.50

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.000	0.024	0.8776
Season	3	0.178	3.367	0.4116
Cow within farm	6	0.565	5.339	0.0025
Farm*Season	3	0.297	5.615	0.0068
Season*Cow within farm	18	0.318		
Duplicate Analysis error	32	0.588		

Table 36. Summary of Data and Analysis of Variance for Cadmium in Milk

Season	Test Farm No. of Samples**	Test Farm Mean (ug/100 ml)	Control Farm No. of Samples**	Control Farm Mean (ug/100 ml)
Fall	3	.30	4	.43
Winter	6	.42	4	.40
Spring	2	.30	1	.30
Summer	1	.20	5	.34

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.036	0.180	0.6764
Season	3	0.973	1.607	0.2228
Cow within farm	6	1.068	0.882	0.5278
Farm*Season	3	0.602	0.994	0.4181
Season*Cow within farm	18	3.632		
Duplicate analysis error	32	2.093		

**Samples below lower detectable limit excluded; analysis of variance based on maximum values (i.e. 0.2 ug/100 ml) for sample tests that were below lower detectable limits.

Table 37. Summary of Data and Analysis of Variance for Copper in Milk

Season	Test Farm No. of Samples**	Test Farm Mean (ug/100 ml)	Control Farm No. of Samples**	Control Farm Mean (ug/100 ml)
Fall	4	13.50	5	8.66
Winter	3	6.43	6	10.07
Spring	8	8.48	8	10.28
Summer	1	9.00	4	11.50

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.196	2.296	0.1470
Season	3	0.204	0.795	0.5127
Cow within farm	6	1.588	3.096	0.0291
Farm*Season	3	0.578	2.253	0.1171
Season*Cow within farm	18	1.539		
Duplicate analysis error	32	1.177		

**Samples below lower detectable limit excluded; analysis of variance based on maximum values (i.e. 7.0 ug/100 ml) for sample tests that were below lower detectable limits.

Table 38. Summary of Data and Analysis of Variance for Lead in Milk

Season	Test Farm No. of Samples**	Test Farm Mean (ug/100 ml)	Control Farm No. of Samples**	Control Farm Mean (ug/100 ml)
Fall	4	3.50	8	13.00
Winter	8	20.25	8	8.00
Spring	8	25.00	7	6.86
Summer	7	5.71	0	

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	2.662	9.924	0.0055
Season	3	22.266	27.666	0.0000
Cow within farm	6	1.492	0.927	0.4992
Farm*Season	3	22.630	28.118	0.0000
Season*Cow within farm	18	4.829		
Duplicate analysis error	32	5.028		

**Samples below lower detectable limit excluded; analysis of variance based on maximum values (i.e. 2.0ug/100 ml) for sample tests that were below lower detectable limit.

Table 39. Summary of Data and Analysis of Variance for Zinc in Milk

Season	Test Farm No. of Samples	Test Farm Mean (ug/100 ml)	Control Farm No. of Samples	Control Farm Mean (ug/100 ml)
Fall	8	193.38	8	280.00
Winter	8	339.25	8	345.88
Spring	8	275.75	8	375.88
Summer	8	313.75	8	343.75

Analysis of Variance

Source	d.f.	Sum of Squares	F	p
Farm	1	0.843	2.835	0.1095
Season	3	2.577	2.889	0.0641
Cow within farm	6	3.150	1.765	0.1633
Farm*Season	3	0.753	0.844	0.4877
Season*Cow within farm	18	5.354		
Duplicate analysis error	32	0.391		

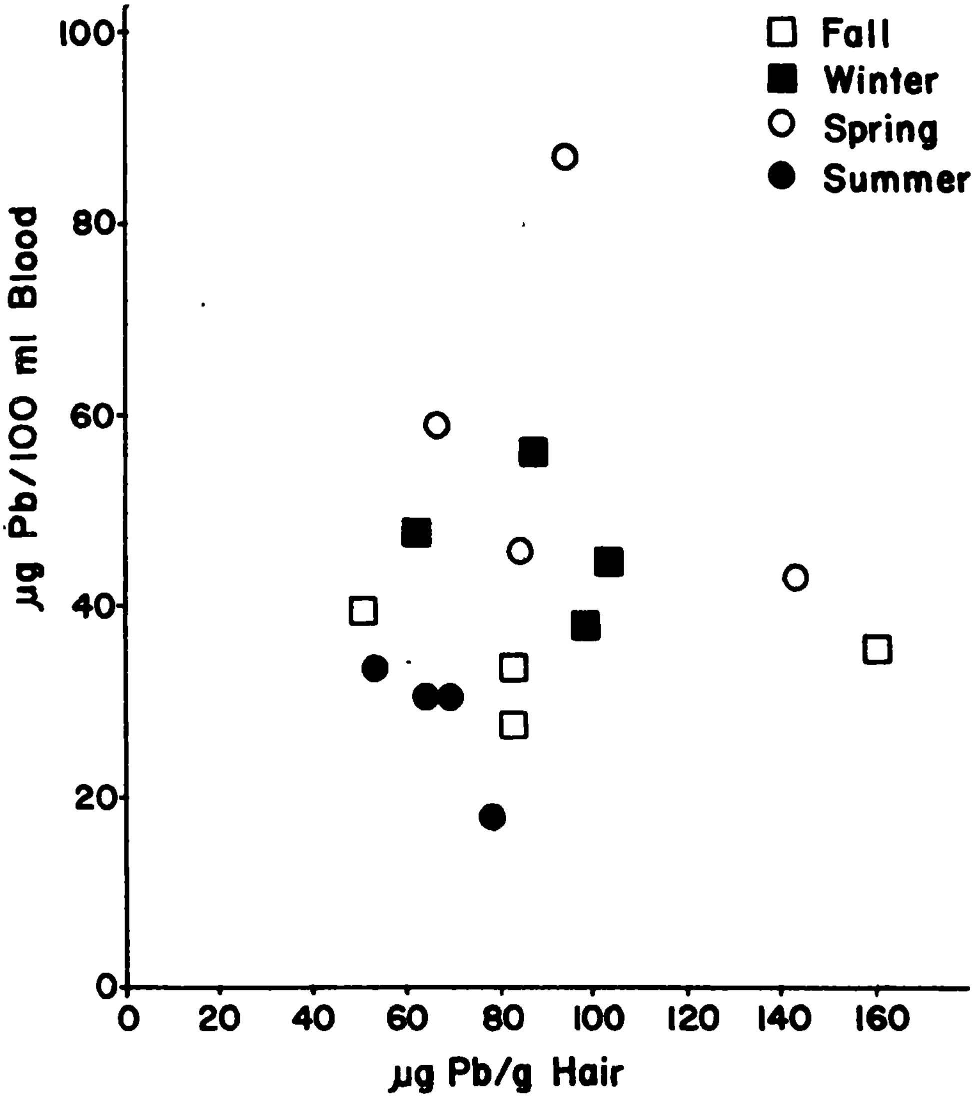

Figure 2. Relationship Between Lead Concentrations in Blood and Hair of Cows on Test Farm

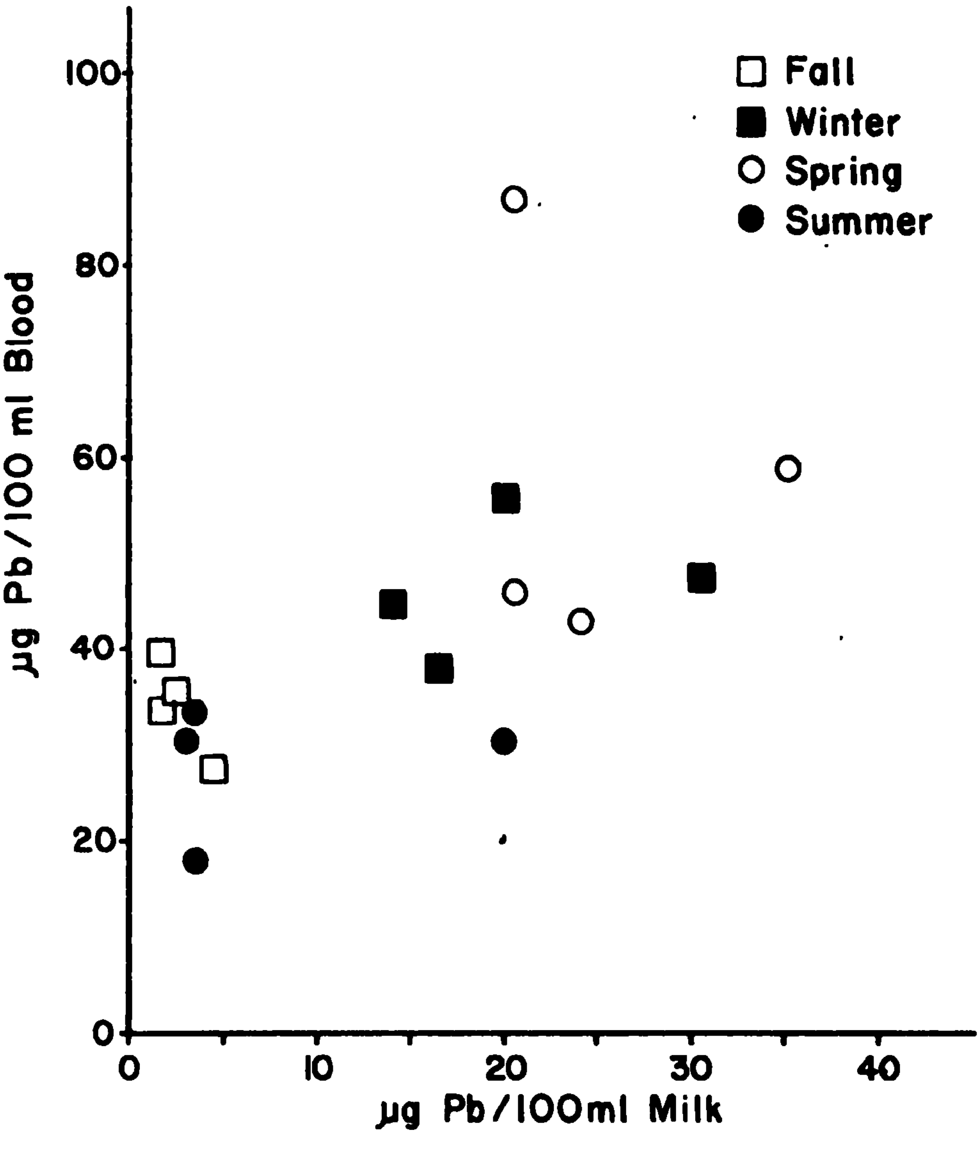

Figure 3. Relationship Between Lead Concentrations in Blood and Milk of Cows on Test Farm

H. Cattle Tissues

Lead was found in higher concentrations in the liver, kidney cortex, muscle and bone of the project owned test cow than in corresponding tissues of the project owned control cow (Table 40). Cadmium was also higher in liver and especially the kidney cortex of the test cow than the control cow. Zinc was higher in liver and bone of the control cow than in the liver and bone of the test cow, and copper was higher in all tissues tested from the control cow. There were no large differences between levels of each of the 4 metals in the muscle samples from the test and control cows. The highest concentration of Cd was in the kidney cortex; copper was highest in liver; and Pb and Zn were highest in blood. The test cow had approximately 17 times higher level of Pb in her liver than the control cow and almost 16 times more Pb in her kidney cortex than the control cow.

I. Human Consumption of Meat and Milk from Cows Exposed to Production Sources of Heavy Metals

None of the cows, on either the test or control farms became ill, so they represent heavy metal exposed cattle that would pass routine inspection and enter meat and milk supplies. Schroeder and Tipton estimated that the lead intake averages 100-500 ug/day, assuming a person consumes about 2,000 gm of food and drink per day.[16] Questions that might be asked are: Would substitution of meat and milk from high metal exposed cows for "normal" meat and milk be harmful

Table 40. Results of Cd, Cu, Pb and Zn Analyses* of Tissues Collected at End of Study Period from Genetically Related Cows on Test and Control Farms

Element and tissue	Mean value (ug/gm)*	
	Test Cow	Control Cow
<u>Cadmium</u>		
Liver	0.90	0.24
Kidney cortex	3.70	1.40
Muscle**	0.10+	0.10+
Bone	<0.05	<0.05
<u>Copper</u>		
Liver	7.25	60.00
Kidney cortex	2.75	3.90
Muscle**	1.30	1.50
Bone	0.58	0.74
<u>Lead</u>		
Liver	2.35	0.14
Kidney cortex	3.75	0.24
Muscle**	0.19	0.06
Bone	9.00	7.1
<u>Zinc</u>		
Liver	33.35	51.6
Kidney cortex	17.55	19.6
Muscle**	43.50	41.9
Bone	70.05	88.5

*Mean value of 2 duplicate samples

**Diaphragm

+One sample below lower detectable limit (0.10 ug/gm) to some persons? If it is harmful, under what circumstances will harm result? Assuming that a growing boy, 8-10 years of age, eats 6 oz of meat and drinks 1 qt of milk per day, the consumption of meat and milk from the test cow rather than the control cow would result in an added 123 ug lead per day. Before this excess dietary lead could result in toxicity, it would be necessary for a person to have considerable exposure to other sources and a resultant high body burden. Other human health effects of lead such as neurological changes, however, may result from chronic exposure to subtoxic doses.

A comparison of the meat and milk Pb values with those for garden crops provides some perspective for evaluation of the dietary implications. Kehoe _et al_. found lead in practically all food items tested including samples from a primitive region far from industrial and mining activities.[17] Leaf and root vegetables are usually higher than kernel and other vegetables.[1] Because of the variety of food items that make up the diet, at the present time food items such as vegetables probably contribute more lead than the meat and milk components. It is, however, disturbing that currently the lead content of some food is considerably higher than in the past.

Even though meat and milk from exposed cattle may not presently constitute a general risk for the consumer, reasonable tolerances would help to both protect the consumer against extreme situations and provide an incentive for the affected businesses to end the mounting lead contamination of the environment. For example, the lead concentration in soil at the 60 ft site increased 219%, 140 ft site increased 257% and 220 ft site increased 284% during a 9 month period on the test farm. The soil there already is approximately 10 times higher in lead than control farm soil. Natural lead removal processes are grossly inadequate to balance the amount of lead being deposited on the soil. The build up of lead has been fairly rapid because the smelter at Glover first started production in 1968. Extensive trucking of ore from the mines in Clark National Forest started at that same time.

The only restrictions on lead in foods in the United States are Food and Drug Administration tolerances for fruit, contained in regulations which were enacted to protect against the effects of improper use of lead arsenate orchard sprays.[18] These tolerances range from 1 ppm for citrus to 7 ppm for apples, apricots and tomatoes. Great Britain has adopted lead tolerances in foods and Canada has proposed similar standards.[19-21] Their tolerance for lead in liver is 2 ppm and fish and edible bone meal is 10 ppm. The British standard permits up to 0.2 ppm in milk. Under the British and Canadian standards, both the slaughtered test cow's liver and kidneys Pb concentrations would have exceeded the tolerance limit.* One test cow's milk

*In an earlier progress report, dated September 19, 1972, it was stated that all specimens were under British regulations for beef products. Since that time it has been determined that the Lead in Food Regulations of 1961 established the limit of 2.0 ppm in liver. The 5.0 ppm limit in the Regulation apparently applies to canned meat products and meat extracts.

sample collected in January and all 4 test cows' milk samples collected in April exceeded the British milk standard of 0.2 ppm. All of the other samples from the test farm cows, the control cows and a cow located on a farm 7.6 miles west from Glover on State Highway 72/21 (thus primarily exposed to ore spillage instead of smelter sources of heavy metal contamination) were below the British milk standards.

J. Garden Crops

A few samples of garden crops raised in the vicinity of the test and control farms were also collected (Table 41). The test area samples were collected from a farm approximately 2 miles north of the test farm. The available gardens did not have lettuce or root vegetables so the samples tested were green beans, bell peppers and tomatoes. The levels of Cd, Cu, Pb and Zn were generally low and there were no differences between the test and control farms. Other garden samples collected in the Glover area have contained higher than normal levels of lead.[22]

K. Lead Translocation

The concentrations of lead at each level of the food chain were compared to derive some estimates that might be used in evaluating the consequences of increased environmental contamination on the lead levels in meat and milk produced in an area. A translocation model was developed for the components of the ecosystem under study, and corresponding lead concentrations were abstracted from the data for the test farm (Figure 4) and

Table 41. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Garden Crop Samples Collected in the Vicinity of the Test and Control Farms, July 28, 1972

Location & Item	Element (ug/gm)			
	Cd	Cu	Pb	Zn
Test Area				
Green Beans	<.5	10.4	5.3	33.5
Bell Pepper	<.5	10.3	<5.0	30.0
Tomato	.75	9.2	5.0	28.2
Control Area				
Green Beans	<.5	9.4	<5.0	40.5
Bell Pepper	<.5	11.5	<5.0	28.2
Tomato	<.5	5.8	<5.0	17.0

the control farm (Figure 5). The entire study period (i.e. 4 samplings) was used for determining the lead levels in air, soil, vegetation, and water that would have contributed to the cows' body burden and the levels measured in hair, blood, milk, liver, kidney cortex, muscle and bone of the cows slaughtered at the end of the study period. The 220 ft samples were used in these calculations because they represented, better than the 60 ft and 140 ft samples, the overall levels that would contribute to the cow's body burden. Additional samples of soil and vegetation were collected on both the test and control farms in October 1971 and July 1972 at 440 ft from the highway. These samples, from near the centers of the fields, were very close to the 220 ft values thus indicating that the 220 ft site was far enough from the highway to represent the general level in the pasture.

From the data presented in Figures 4 and 5, it can be seen that a difference between dustfall Pb on test and control farms of approximately 10 fold, corresponded approximately to 3.2 times higher muscle Pb concentration and a 5 times higher milk Pb concentration for the slaughtered test cow than for the slaughtered control cow. A similar relationship was found when suspended lead values from the air filter data were used. The mean of the suspended lead values (2.1671 ug/m^3) on the test farm for the sampling periods was approximately 15 times higher than the mean of the sampling periods on the control farm (0.1431 ug/m^3).

Soil samples would be more easily obtained than dustfall

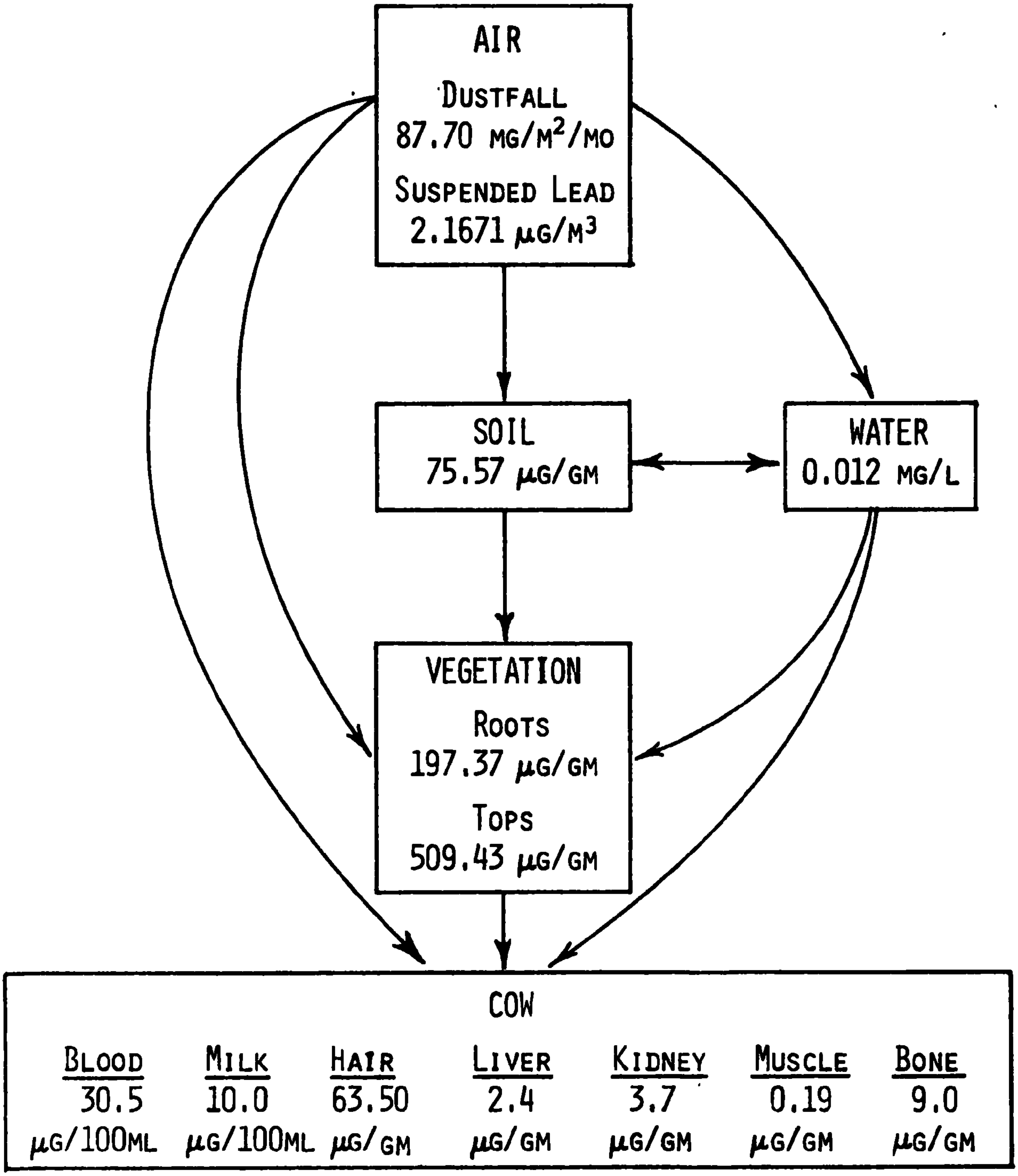

Figure 4. Test Farm Lead Translocation Model

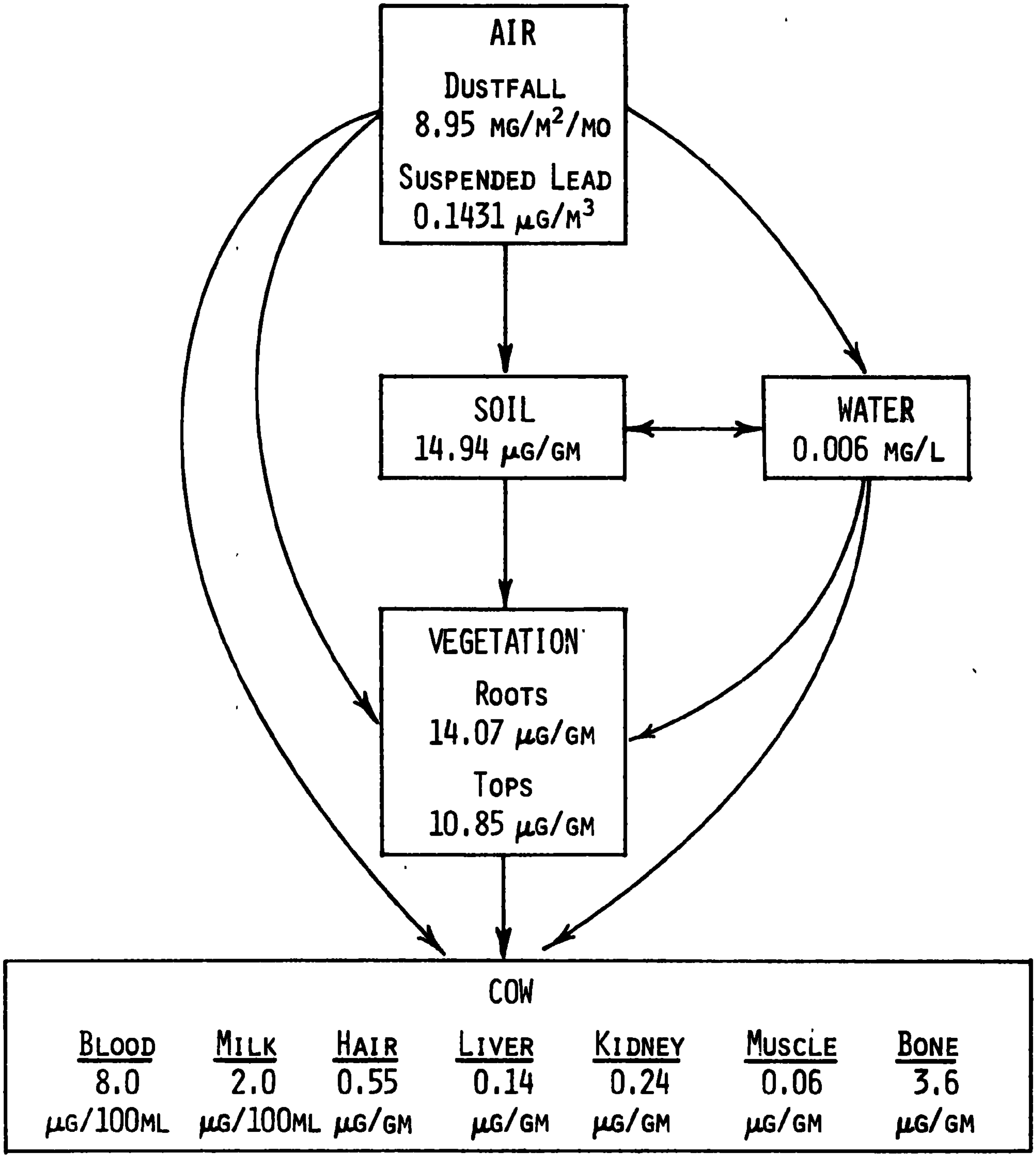

Figure 5. Control Farm Lead Translocation Model

or air filter samples for predicting the muscle and milk lead levels of cattle grazing on the soil. The 5 times higher mean soil Pb concentration for all samplings on the test farm than on the control farm corresponded to the previously described 3.2 times higher muscle Pb and 5 times higher milk Pb on the test farm than on the control farm. On an equivalent basis, the test cow's blood Pb concentration was 1/246 the test farm soil concentration and the control cow's blood Pb concentration was 1/187 the control farm soil concentration. This relationship should not, however, be expected to be the same in other situations involving markedly different Pb contamination sources. For example, in the models developed for the test and control farm, water appeared to have very little contribution to the cattle's intake of lead. The pH of the soil, type of vegetation, use of fertilizers and lime, rainfall and other meteorological conditions would also affect the relationships between air and soil lead levels and the levels found in animal food products for human consumption produced in a given area.

On the control farm, there was a general dilution in the lead concentrations at each step in transport between soil and biological tissues (Figure 4). The roots had a lower concentration than soil, unwashed vegetation tops lower than roots, and all body tissues were lower than the tops. The bone concentration of lead was higher than other body tissues sampled and milk was less than 1/4 the blood lead level.

On the test farm, a different pattern was observed. The concentration of lead in roots was 2.6 times higher than soil, unwashed vegetation tops were 2.6 times higher than roots, and finally all body tissues were less than the tops. Milk had between 1/3 and 1/4 the concentration of lead found in the blood.

The high lead levels for unwashed vegetation on the test farm obviously reflected the airborne contamination because the washed vegetation tops were only 84.5% the level for unwashed vegetation tops (i.e. the washings contained 79 ug Pb/gm or accounted for 15.5% of the unwashed tops value). The consistent findings in all seasons of higher lead concentrations in roots than in soil on the test farm, but not on the control farm, indicates that under these conditions of relatively high levels of contamination, there was bio-magnification of lead in the roots. These observations also lend support to the view that much of the lead in vegetation, foliage and roots, enters the plant by foliar absorption.[23-24]

It is commonly thought that in an area of lead contamination, that horses are more likely to develop lead poisoning than cattle grazing on the same pasture.[25-27] In fact, that relationship has been observed in the Glover area as documented horse deaths due to lead poisoning occurred, but no cattle deaths have been diagnosed as lead poisoning. It has been suggested that the grazing habits of the horse may be a reason for the greater apparent susceptibility of the horse, versus the cow, to lead poisoning.[28] Horses occasionally pull forage out by the roots and eat the roots and attendant soil along with the forage. The data reported here show that the soil and root lead concentrations are much less than unwashed vegetation tops so a horse would have less intake if the roots substituted for some of the tops in its diet. Therefore, it seems that the basis for observations of horses being afflicted more than cattle by lead poisoning in an area of contamination is for other reasons than their grazing habits, probably a lower biological tolerance on the part of the horse.

L. Intake and Assimilation of Lead by Cattle

Of the total lead intake, only a portion is assimilated by the body. Assimilation occurs in two ways: absorption from the alimentary tract and absorption from the respiratory tract. Lead transported via these 2 pathways contribute to the body burden, and each pathway will be considered separately to facilitate the determination of the total amount of lead assimilated.

1. Lead intake from the diet

An estimate of the daily ration of a cow can be arrived at by knowing its nutritional needs and the nutritional value of the food available to the cow for consumption. The basis used for determining the daily nutritional requirement was megacalories of digestible energy, which in turn is dependent on body weight, milk production, butterfat content of the milk and period of gestation. From the foodstuffs available and their dates of feeding, a ration was calculated that was adequate in fulfilling the cow's daily requirement for energy.

At the test farm, the ration consisted of ground corn and hay or pasture forage depending on the season of the year, i.e. hay in the winter and pasture forage the rest of the year. At the control farm, the ration was essentially the same except that corn silage was offered along with hay in the winter.

During each sampling period, grain and vegetation samples were collected. Hay and silage samples were obtained during the winter and spring sampling. Laboratory results were reported as ug/gm; therefore, by knowing the amounts of grain, hay, silage, or pasture forage the cow consumes, it was possible to calculate the total amount of lead ingested. The daily intakes of the project owned cows on the test farm and on the control farm were calculated from the ration and the nutritive requirements of each cow.[29] It was estimated that, depending on the season, the test cow consumed 18-21 kg feed per day and the control cow consumes 26-32 kg feed per day. Assuming a cow drinks 29 gallons of water per day, the intake of lead from water on the test farm (0.012 mg/l) and control farm (<0.006 mg/l) was negligible. Based upon feed intake, the test cow was consuming 8.65 mg Pb per kg of body weight, or 4762.92 mg Pb per day, and the control cow was consuming 0.78 mg Pb per kg of body weight, or 430.54 mg Pb per day, over the entire year. The specific values for the 4 seasons are shown in Table 42. Based upon radioisotope studies, one percent of this dietary intake was assumed to be absorbed by the body.[30]

2. Lead intake from respiration

An estimate of the amount of lead inhaled per day by a cow can be computed if the average concentration of airborne suspended lead is known. Determination of suspended lead was accomplished by continuous air sampling during 4 to 6 weeks of the 13 weeks in a sampling period. Suspended lead in air on the test farm remained relatively constant during the fall, spring and summer months (Table 43). The winter period, however, had a level 2 times greater than the mean value for all seasons on the test farm. At the control farm, the level of suspended lead declined steadily throughout the year with the last period being less than 50% of the first period. Overall, the test farm had higher suspended lead levels than the control farm with the test/control ratio for lead being 16.5. On the control farm, lead concentration in air was assumed to be stable at the location of air sampling approximately 400 meters from the highway and the levels found in the first 4 weeks of each sampling period.

Multiplying the lead concentrations in ambient air by

Table 42. Daily Dietary Intake and Respiratory Intake of Lead by Project Owned Test Cow and Control Cow by Season

	Test Cow		Control Cow	
Season	mg Pb/kg body wt/day	mg Pb/day	mg Pb/kg body wt/day	mg Pb/day
Dietary intake*				
All seasons	8.65	4762.92	0.78	430.54
Fall	7.65	4205.40	0.88	486.44
Winter	10.05	5529.28	0.71	388.59
Spring	14.13	7772.73	0.95	520.11
Summer	2.77	1524.04	0.59	327.02
Respiratory intake+				
All seasons	0.00057	0.3126	0.00004	0.0207
Fall	0.00045	0.2464	0.00006	0.0320
Winter	0.00110	0.6050	0.00004	0.0201
Spring	0.00036	0.1978	0.00003	0.0179
Summer	0.00037	0.2011	0.00002	0.0127

*Assumption: 550 kg body weight

+Assumption: 156.9942 m^3/day inhaled air

Table 43. Comparison of Suspended* Cd, Cu, Pb and Zn on Test and Control Farms

Season**	Test Farm (ug/m^3)+				Control Farm (ug/m^3)+			
	Cd	Cu	Pb	Zn	Cd	Cu	Pb	Zn
All seasons	.0259	.0282	2.1474	.5141	.0023	.0068	.1304	.4111
Fall	.0316	.0278	1.5692	.7261	.0051	.0057	.2037	.5754
Winter	.0420	.0404	3.8536	.7601	.0014	.0130	.1282	.8384
Spring	.0137	.0274	1.5507	.1725	.0013	.0041	.1068	.0141
Summer	.0130	.0147	1.3001	.3446	.0015	.0028	.0804	.1127

*Gelman Glass Fiber filters

**Approximately 4 week sampling period for each season; the season total is a weighted average of the season-specific values.

+Values corrected by subtracting blank value from the sample measurement.

the cows' total respiratory volume in a day (156.9942 m^3; from Brody[31]) would result in the amount of lead inhaled by cows each day. For the purposes of estimation, it was assumed that the cow was similar to man with 30% pulmonary retention of inhaled lead. The absolute amount of lead via the respiratory tract was only 0.19% as great as the amount of dietary assimilation on the test farm and 0.13% as great as the amount of dietary assimilation on the control farm.

3. Total lead assimilation

By combining the amount of lead absorption from food and water and the amount retained from inhaled air, it was determined that the test cow was assimilating during the season of highest exposure (spring), approximately 0.14 mg/kg body weight and the control cow was receiving approximately 0.01 mg/kg body weight during the same season. Hammond and Aronson[25] have estimated that the minimum cumulative lethal dose for a cow is 6-7 mg/kg body weight, and Allcroft[32] succeeded in producing a chronic syndrome and death after 33 months of continuous feeding of 5-6 mg Pb/kg/day to a steer. The mean seasonal daily intake calculated in the present study for the test cow (Table 42) was above these dosages in all of the seasons except summer, but the cow was never observed to be ill or exhibit signs of lead toxicity. This is not necessarily in conflict with the earlier observations as this cow was 10 years old.

The minimum cumulative lethal dose would be expected to be higher for older cows, as younger animals are usually more sensitive to lead.

4. Relationship between total lead assimilation and blood lead

Based upon data collected by Kehoe, the relationship between total lead assimilated from both the gastrointestinal tract and lungs of man were compared with the subjects blood levels.[33] Allcroft[32] and Allcroft and Blaxter[34] have published results of cattle experiments in which the blood lead concentrations were determined for various amounts of lead fed in the ration. It was assumed in these studies that the contribution of lead in inhaled air was negligible. The present study provided an opportunity to examine under natural conditions the relationship between total lead absorption in the cow, from both ingested and inhaled lead, and the blood lead levels.

As shown in Figure 6, there is a peak in assimilated lead during the spring period on both farms with the test cow being approximately 15 times higher than the control cow. The blood lead levels also peaked at this time with the level of the cow on the test farm being almost 5 times greater than the level of the cow on the control farm. The milk lead level of the test cow also reflected this increase of lead intake by increasing and peaking during this period, although the lead level in the control cow's milk did not peak and continued to decline. Overall, the mean blood lead value of the cow on the test farm was 3.8 times greater than the mean blood lead value of the cow on the control farm, and the mean milk lead value of the cow on the test farm was 1.6 times greater than the mean milk lead value of the cow on the control farm.

M. Meteorological Effects and Sources of Contamination on the Test Farm

Wind data were collected at the test farm for a total of 331 days over a 366 day period. Mechanical break-down of recorder and occasionaly lack of strip-chart paper accounted for 35 days for which no data was collected. The main 3 wind directions and their corresponding mean velocities were South (26.59%) at 4.4 mph, Southwest (24.17%) at 2.2 mph and North (23.87%) at 5.6 mph.

Tables 44 and 45 contain seasonal data for suspended Cd and Pb levels in air, vehicular traffic (for the previous year) wind direction and wind velocity at the test farm. It is north of the smelter and north of State Highway 72/21 so wind direction and velocity would affect in a similar manner the recognized sources of heavy metal contamination, namely vehicular emissions, smelter stack emissions (the stack is 610 ft tall), ore truck spillage, and dust from stockpiled ore on the smelter premises. Assuming that the seasonal pattern of vehicular traffic by the farm during the study period was similar to the preceding year, there was a negative relationship with the levels of Cd and Pb in the air. In the winter and spring when the suspended Cd and Pb levels were highest in the air, the traffic was lowest. Therefore, it appears that the contribution by vehicular emissions has a minor role in determining the airborne Cd and Pb levels. It is not known if the ore truck traffic corresponds to the total vehicular traffic pattern. The trucking of ore to the smelter operated independently of the smelter operating schedule because the ore was stockpiled to ensure a constant supply when the smelter sinter and blast furnaces were operating. Due to the hilly terrain and adverse driving conditions in the winter, it is thought that the amount of trucking was less in the winter than in other seasons. A more important factor is wetting the lead ore loaded onto the trucks to avoid dust and covering of the two ore hoppers on each truck to avoid spillage. It is not known if these two procedures were performed more diligently in some seasons than others; however, the project investigators have personally seen uncovered ore trucks on State Highway 72/21 during all 4 seasons.

The smelter is an obvious major source of airborne lead in the area. During 1971, the smelter production was erratic because of a labor strike. The strike was ended in Feb., 1971 and the numbers of days that the sinter and blast furnace were operated during each of the previously described sampling periods in this study were consistent.

The wind, as determined by the weather station maintained on the test farm, was from a quadrant (SE,S,SW) that would blow the smelter plume and any dust from the property towards the test farm more of the time during the winter and summer periods than other periods. Conversely, the wind was observed to blow towards the smelter, away from the farm, much of the time during the fall and spring study periods. Therefore, it appeared that the southerly wind had a major effect, increasing the levels of suspended Pb and Cd on the test farm. The wind velocity had variable effect on the levels of suspended Pb and Cd. The hypothesis that the main source of the contamination on the test farm is the smelter stack emissions, is supported by the 10 fold higher dustfall Pb at the 220 ft site and 13 fold higher airborne suspended Pb on the test farm as compared with corresponding measurements on the control farm. It is unlikely that the main highway sources (engine emissions and ore spillage) would contribute much to the 220 ft measurements. Furthermore, the gradient had leveled off at the 220 ft site to values that were similar to the general levels found near the center of the field at the 440 ft collection location. The distance between the test farm and the smelter property, approximately 800 meters or 0.5 mile, would greatly diminish the amount of surface wind distribution of stockpiled ore to the test farm during times when the wind was blowing from a southern quadrant.

It was difficult to fully evaluate the contribution of spillage from ore trucks to the levels of lead in the cattle on the test farm, so another cow from a farm on the north side of State Highway 72/21 approximately 7.6 miles west of Glover was sampled in the fall, spring and summer (Table 46). The mean concentrations of lead in both blood and milk of the cow exposed to ore spillage (plus background due primarily to vehicular emissions but not smelter emissions) were intermediate between that of the cows exposed to multiple sources on the test farm and cows exposed to only background on the control farm. The milk lead mean concentration of the ore spillage exposed cow (11.8 ug/100 ml) was almost as large as the milk concentrations of the cows on the test farm.

Table 44. Settleable and Suspended Cadmium (Cd) by Season, Vehicular Traffic, and Wind Direction and Velocity.

Measurement	Samples per season	Season: Fall	Winter	Spring	Summer
Dustfall Cd[1] ($mg/m^2/mo.$)					
60'	3	0.8716*	1.8316	0.5954+	1.6082
140'	3	0.8680	1.6824	0.5589+	1.3249
220'	3	0.5466	1.4711	0.4825	1.2407
Total Suspended Cd[2] (ug/m^3)	3	0.0316	0.0420	0.0106	0.0209
Vehicular Traffic[3]		76077	58706	85107	96196
		Percent			
Wind Direction		100.00	100.00	100.00	100.00
Toward Farm (SE,S,SW)		39.58	59.30	49.45	60.44
Not Toward Farm		60.42	40.70	50.55	39.56
		Percent			
Wind Velocity[4]		100.00	100.00	100.00	100.00
0-5 mph		66.67	51.52	66.67	72.53
5-10 mph		29.88	39.39	30.95	26.38
10-15 mph		3.45	9.09	2.38	1.09

*One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

+One sample lost during lab preparation; therefore, the value presented is based on two samples.

[1]Fall=Oct.2,1971-Jan.6,1972; Winter=Jan.6,1972-Apr.1,1972; Spring=Apr.1,1972-Jul.1,1972; Summer=Jul.1,1972-Oct.1,1972.

[2]Weighted average based on percent of total collection period that each filter was used. Fall=Oct.3,1971-Oct.30,1971; Winter=Jan.6,1972-Feb.6,1972; Spring=Apr.1,1972-Apr.28,1972 and May 5,1972-May 19,1972; Summer=Jul.1,1972-Jul.28,1972 and Aug.4,1972-Aug.20,1972.

[3]Factored average traffic volume for 1970-1971 time periods equivalent to footnote 1; from Missouri State Highway Commission data.

[4]Average velocity of daily-prevailing-winds.

Table 45. Settleable and Suspended Lead (Pb) by Season, Vehicular Traffic, and Wind Direction and Velocity.

Measurement	Samples per season	Season: Fall	Winter	Spring	Summer
Dustfall Pb[1]					
(mg/m^2/mo.)					
60'	3	85.9698	170.3140	100.5931	121.9600
140'	3	130.4618	130.9470	87.0318	89.0289
220'	3	76.4983	122.9700	72.7931	78.4986
Total Suspended Pb[2]					
(ug/m^3)	3	1.5692	3.8536	1.2599	1.2810
Vehicular Traffic[3]		76077	58706	85107	96196
		Percent			
Wind Direction		100.00	100.00	100.00	100.00
Toward Farm (SE,S,SW)		39.58	59.30	49.45	60.44
Not Toward Farm		60.42	40.70	50.55	39.56
		Percent			
Wind Velocity[4]		100.00	100.00	100.00	100.00
0-5 mph		66.67	51.52	66.67	72.53
5-10 mph		29.88	39.39	30.95	26.38
10-15 mph		3.45	9.09	2.38	1.09

[1]Fall=Oct. 2, 1971-Jan. 6, 1972; Winter=Jan. 6, 1972-Apr. 1, 1972; Spring= Apr. 1, 1972-July 1, 1972; Summer=July 1, 1972-Oct. 1, 1972.

[2]Weighted average based on percent of total collection period that each filter was used. Fall=Oct. 3, 1971-Oct. 30, 1971; Winter=Jan. 6, 1972-Feb. 6, 1972; Spring=Apr. 1, 1972-Apr. 28, 1972; and May 5, 1972-May 19, 1972; Summer=July 1, 1972-July 28, 1972 and Aug. 4, 1972-Aug. 20, 1972.

[3]Factored average traffic volume for 1970-1971 time periods equivalent to footnote 1; from Missouri State Highway Commission data.

[4]Average velocity of daily-prevailing-winds.

Table 46. Comparison of Mean Bovine Blood and Milk Lead Values by Season and Location of Farm

Specimen and Season	Test Farm		Farm 7.6 Miles West of Glover		Control Farm	
	No. of Samples	ug/100 ml	No. of samples	ug/100 ml	No. of Samples	ug/100 ml
Blood						
All seasons	32	41.8	4	17.5	32	11.2
Fall	8	34.0	2	22.0	8	18.9
Winter	8	46.5	-	-	8	8.8
Spring	8	58.8	1	8.0	8	10.4
Summer	8	28.1	1	18.0	8	6.6
Milk						
All seasons	32	13.0	4	11.8	32	6.8
Fall	8	1.8	2	16.5	8	13.0
Winter	8	20.3	-	-	8	8.0
Spring	8	25.0	1	14.0	8	6.0
Summer	8	5.0	1	<2.0	8	<2.0

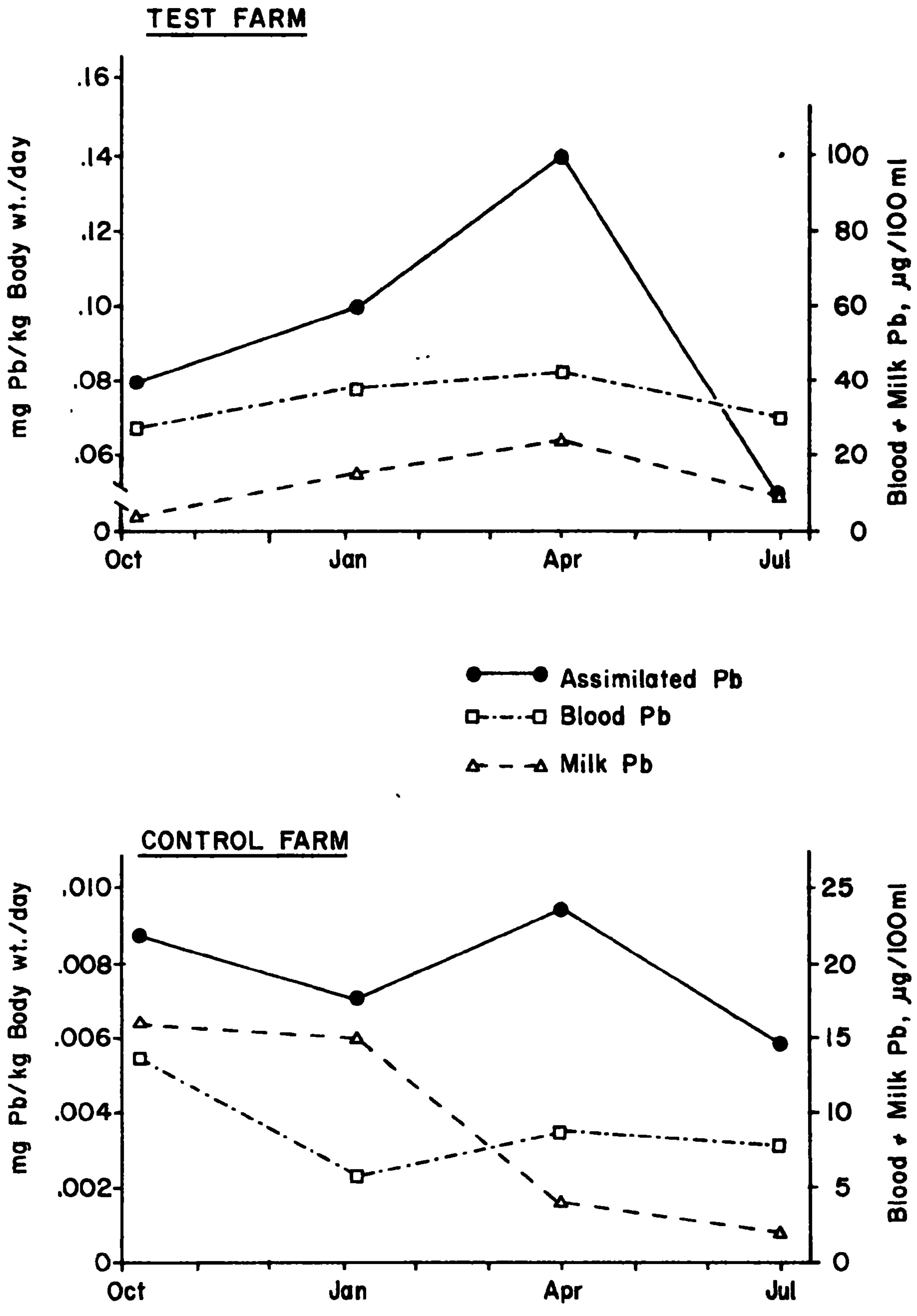

Figure 6. Total Assimilated Lead, Blood Lead, and Milk Lead for Project Owned Test Cow and Control Cow

VI.

REFERENCES

VI. REFERENCES

1. Committee on Biological Effects of Atmospheric Pollutants: Airborne Lead in Perspective. National Academy of Sciences, Wash. D.C., 1972.

2. Kobayashi, J.: Relation between the "Itai-Itai" disease and the pollution of river water by cadmium from a mine. Proc. 5th International Water Pollution Research Conference, San Francisco, July-August, 1970.

3. Underwood, E.J.: Trace elements in human and animal nutrition. Academic Press, New York, 1956.

4. Chow, T.J. and Patterson, C.C.: The occurrence and significance of lead isotopes in pelagic sediments. Geochim. Cosmochim. Acta 26:263-308, 1962.

5. Missouri Geological Survey and Water Resources: The World's No. 1 Lead Producer. Missouri Mineral News, Rolla, Mo. Vol. 11, No. 3, 1971.

6. Gibson, F.W.: New Buick lead smelter incorporates forty years of technical advances. Engineering and Mining Journal, 1968.

7. Wixson, B.G., and Bolter, E.: Evaluations of stream pollution and trace substances in the new lead belt of Missouri. Proc. 5th Annual Conf. on Trace Substances in Environmental Health, University of Missouri, Columbia, 1972.

8. Connor, J.J., Erdman, J.A., Sim, J.D., and Ebens, R.J.: Roadside effects on trace element content of some rocks, soils and plants of Missouri. <u>In</u> Hemphill, D.D. (ed.) Proc. 4th Annual Conf. Trace Subst. in Environ. Health. Univ. of Missouri, Columbia, Mo. 1970.

9. Pierce, J.O. and Cholak, J.: Lead, chromium and molybdenum by atomic absorption. Arch. Environ. Health 13:208-212, 1966.

10. Analytical Methods for Atomic Absorption Spectrophotometry. Perkin-Elmer Corp., Norwalk, Conn., 1968.

11. Pickett, E.E.: Current capabilities in analysis of trace substances: flame photometry and atomic absorption. Proc. 1st Annual Conf. Trace Substances in Environmental Health. Univ. of Missouri, Columbia, 1967.

12. Kirtyohann, S.R. and Pickett, E.E.: Spectral interferences in atomic absorption spectrophotometry. Anal. Chem. 38:585-587, 1966.

13. Mitchell, R.L. and Reith, J.W.S.: The lead content of pasture herbage. J. Sci. Food Agric. 17:437-440, 1966.

14. Everett, J.C., Day, C.L. and Reynolds, D.: Comparative survey of lead at selected sites in the British Isles in relation to air pollution. Food Cosmet. Toxicol. 5:29-35, 1967.

15. Nishujama, K. and Nordberg, G.F.: Adsorption and elution of cadmium on hair. Arch. Environ. Health 25:92-96, 1972.

16. Shroeder, H.A. and Tipton, I.H.: The human body burden of lead. Arch. Environ. Health 17:965-978, 1968.

17. Kehoe, R.A., Thamann, F. and Cholak, J.: On the normal absorption and excretion of lead. I. Lead absorption and excretion in primitive life. J. Ind. Hyg. 15:257-272, 1933.

18. Food and Drug Administration: Title 21 Food and Drugs, Section 120.194. Code of Federal Regulations, U.S. Govern. Printing Office.

19. Ministry of Agriculture, Fisheries and Food and Ministry of Health: The Lead in Food Regulation, 1961. Food and Drugs Composition, England and Wales, No. 1931. Her Majesty's Stationery Office, 1961.

20. Peden, J.D.: A survey of the arsenic, copper and lead contents of pigs and other animal livers. J. Assoc. Public Analysts 8:14-19, 1970.

21. Anonymous: Lead. Food Chemical News 12:12, Oct. 5, 1970.

22. Hemphill, D.: Personal communication, Nov., 1972.

23. Chamberlain, A.C.: Interception and retention of radioactive aerosols by vegetation. Atmos. Environ. 4:57-78, 1970.

24. Mueller, P.K. and Stanley, R.L.: Origin of lead in surface vegetation. AIHL Report No. 87. State of Calif Dept. of Public Health, Berkeley, Calif.

25. Hammond, P.B. and Aronson, A.L.: Lead poisoning in cattle and horses in the vicinity of a smelter. Annals N.Y. Acad. Sci. Vol. 111, Art. 2, pp. 595-611, 1964.

26. Schmitt, N., Larsen, A.A., McCausland, E.D. and Saville, J.M.: Lead poisoning in horses; an environmental health hazard. Arch. Environ. Health 23:185-195, 1971.

27. California Air Resources Board: A Joint Study of Lead Contamination Relative to Horse Deaths in Southern Solano County. December, 1972.

28. Aronson, A.L.: Lead poisoning in cattle and horses following long-term exposure to lead. Amer. J. Vet. Res. 33:627-629, 1972.

29. National Academy of Science: Nutrient Requirements of Dairy Cattle. National Research Council Publication No. 1349, Third Revised Edition, 1966.

30. Stanley, R.E., Mullen, A.A., and Bretthauer, E.W.: Transfer to milk of ingested radiolead. Health Physics 21:211-215, 1971.
31. Brody, W.: Bioenergetics and Growth. Missouri Agriculture Experiment Station Publication, 1944.
32. Allcroft, R.: Lead as a nutritional hazard to farm livestock. IV. Distribution of lead in the tissues of bovines after ingestion of various lead compounds. J. Comp. Path. 60:190-208, 1950.
33. Kehoe, R.A.: The metabolism of lead in man in health and disease. The Harben Lectures, 1960. J. Roy. Inst. Public Health Hyg. 24:1-81, 101-120, 129-143, 177-203, 1961.
34. Allcroft R. and Blaxter, K.L.: Lead as a nutritional hazard to farm livestock. V. The toxicity of lead to cattle and sheep and an evaluation of the lead hazard under farm conditions. J. Comp. Path. 60:209-218, 1950.

VII.

ACKNOWLEDGMENTS

VII. ACKNOWLEDGMENTS

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VIII.

APPENDIX TABLES

Table 1. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Test Farm near Glover, Missouri During October 2, 1971 - January 6, 1972.

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1] Cd	Cu	Pb	Zn
Blood[2]	1	2	1.60	74.50	35.50	408.50
Blood	2	2	1.50	94.00	33.50	435.00
Blood	3	2	2.00	89.00	27.50	389.00
Blood	4	2	1.85	86.00	39.50	326.50
Milk[2]	1	2	.30*	12.00	2.50	290.50
Milk	2	2	**	**	**	67.00
Milk	3	2	.40*	15.00	4.50	324.00
Milk	4	2	.20*	**	**	92.00
Hair[3]	1	2	1.20	9.10	160.00	118.50
Hair	2	2	.90	8.45	83.00	126.50
Hair	3	2	.95	8.60	83.00	86.00
Hair	4	2	2.10	6.90	50.50	87.00
Soil 60 ft.[3]		3	.58	10.13	71.67	26.67
Soil 140 ft.		3	.53	9.63	43.00	22.00
Soil 220 ft.		3	.56	9.80	41.33	23.67
Roots 60 ft.[3]		3	1.66	23.00	211.33	105.33
Roots 140 ft.		3	1.06	23.00	115.00	60.67
Roots 220 ft.		3	1.90	19.67	298.33	73.67
Vegetation, unwashed[3]						
60 ft.		3	4.18+	14.01	396.00	66.13
140 ft.		3	4.40‡	11.50	296.00	59.40
220 ft.		3	2.96+	11.98	286.00	55.07
Vegetation, washed[3]						
60 ft.		3	2.93	12.13	304.00	58.33
140 ft.		3	2.37	10.23	251.33	55.00
220 ft.		3	2.30	11.03	247.33	52.00
Dustfall 60 ft.[4]		3	.8716+	1.8703	86.0115	1.3546
Dustfall 140 ft.		3	.8680	2.8291	130.4618	1.1621
Dustfall 220 ft.		3	.5466	1.9466	76.5575	1.8268

[1] Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2] Mean value of blood and milk samples as ug/100 ml; Oct. 2, 1971.

[3] Mean value of hair, soil, root and vegetation samples as ug/g; Oct. 2-9, 1971.

[4] Mean value as $mg/m^2/mo$; Oct. 2, 1971 - Jan. 6, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

‡Two sample values were below lower detectable limit; therefore, the value presented is based on one sample.

Table 2. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Control Farm near Ellington, Missouri, During October 2, 1971 - January 4, 1972.

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1] Cd	Cu	Pb	Zn
Blood[2]	1	2	.95	89.50	17.50	520.00
Blood	2	2	4.40	94.50	14.00	456.00
Blood	3	2	1.15	107.00	22.50	481.50
Blood	4	2	2.15	98.00	21.50	523.50
Milk[2]	1	2	**	9.00	12.50	451.00
Milk	2	2	.30*	10.00*	16.00	205.50
Milk	3	2	.60	7.65	9.50	281.50
Milk	4	2	.20*	**	14.00	182.00
Hair[3]	1	2	.06	7.45	2.85	90.50
Hair	2	2	.05	6.50	1.55	94.00
Hair	3	2	.06	7.65	2.40	98.00
Hair	4	2	.07	7.40	1.95	92.50
Soil 60 ft.[3]		3	.41	5.43	13.67	49.67
Soil 140 ft.		3	.42	6.77	13.00	25.00
Soil 220 ft.		3	.38	4.80	12.33	17.00
Roots 60 ft.[3]		3	1.22+	10.63	15.33	259.33
Roots 140 ft.		3	.79	11.67	8.20	36.33
Roots 220 ft.		3	.86	11.67	14.67	78.00
Vegetation, unwashed[3]						
60 ft.		3	**	8.95	21.47	337.37
140 ft.		3	**	13.37	24.97	70.23
220 ft.		3	1.07+	10.06	15.10	65.27
Vegetation, washed[3]						
60 ft.		3	.76	7.97	17.57	309.33
140 ft.		3	.93	10.47	11.70	62.33
220 ft.		3	.69	7.73	10.30	52.33
Dustfall 60 ft.[4]		3	.0356‡	.2373	1.6204	1.5871
Dustfall 140 ft.		3	.3052	1.3090	46.3752	2.7328
Dustfall 220 ft.		3	.2492	.7342	29.7586	1.1310

[1] Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2] Mean value of all blood and milk samples in ug/100 ml; Oct. 2, 1971.

[3] Mean value of hair, soil, root and vegetation samples as ug/g; Oct. 2-3, 1971.

[4] Mean value as $mg/m^2/mo$; Oct. 2, 1971 - Jan. 4, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

‡Two sample values were below lower detectable limit; therefore, the value presented is based on one sample.

Table 3. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Test Farm near Glover, Missouri, During January 6 - March 31, 1972.

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1]			
			Cd	Cu	Pb	Zn
Blood[2]	1	2	.40*	92.00	56.00	440.50
Blood	2	2	.40*	100.00	47.50	361.50
Blood	3	2	.35	112.50	38.00	406.00
Blood	4	2	.40	84.00	44.50	358.00
Milk[2]	1	2	.30	**	20.00	331.50
Milk	2	2	.50	**	30.50	423.00
Milk	3	2	.50*	**	16.50	265.00
Milk	4	2	.40*	8.90*	14.00	337.50
Hair[3]	1	2	1.55	7.25	87.00	195.00
Hair	2	2	1.10	7.85	61.50	128.00
Hair	3	2	2.35	8.65	98.50	109.50
Hair	4	2	1.95	7.30	103.00	107.00
Soil 60 ft.[3]		3	1.06	14.67	122.33	39.20
Soil 140 ft.		3	.79	10.50	49.73	22.60
Soil 220 ft.		3	.88	10.87	61.43	28.93
Roots 60 ft.[3]		3	3.90	32.90	374.67	151.33
Roots 140 ft.		3	2.67	34.47	196.00	84.67
Roots 220 ft.		3	2.83	34.27	145.33	66.33
Vegetation, unwashed[3]						
60 ft.		3	9.72+	23.97	1081.00	885.33
140 ft.		3	8.41+	19.03	1012.33	58.37
220 ft.		3	6.72	16.00	845.00	99.63
Vegetation, washed[3]						
60 ft.		3	7.30	15.33	756.67	48.97
140 ft.		3	7.97	15.70	860.00	47.13
220 ft.		3	5.68	12.87	698.33	89.00
Dustfall 60 ft.[4]		3	1.8316	3.3897	170.3140	18.2331
Dustfall 140 ft.		3	1.6824	2.5775	130.9470	13.1568
Dustfall 220 ft.		3	1.4711	2.2957	122.9700	11.8101

[1]Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2]Mean value of all blood and milk samples in ug/100 ml; Jan. 6, 1972

[3]Mean value of hair, soil, root and vegetation samples as ug/g; Jan. 6, 1972.

[4]Mean value as $mg/m^2/mo$; Jan. 6 - March 31, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

Table 4. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Control Farm near Ellington, Missouri, During January 5 - April 2, 1972.

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1]			
			Cd	Cu	Pb	Zn
Blood[2]	1	2	.35	109.00	13.50	346.50
Blood	2	2	.30*	102.00	6.00	430.50
Blood	3	2	**	121.00	8.00	325.50
Blood	4	2	.50*	112.00	7.50	405.00
Milk[2]	1	2	.55	11.90	8.50	484.00
Milk	2	2	**	7.70	15.00	178.50
Milk	3	2	**	10.60	5.00	357.00
Milk	4	2	.25	**	3.50	364.00
Hair[3]	1	2	.18	9.70	3.95	115.50
Hair	2	2	.06	6.70	1.02	111.00
Hair	3	2	.08	7.55	2.70	117.00
Hair	4	2	.19	7.40	8.00	120.00
Soil 60 ft.[3]		3	.66	6.83	15.47	23.47
Soil 140 ft.		3	.60	8.87	15.23	23.43
Soil 220 ft.		3	.55	7.03	14.70	19.57
Roots 60 ft.[3]		3	.55	18.13	9.50	41.53
Roots 140 ft.		3	.70	20.97	6.80+	46.93
Roots 220 ft.		3	.68	22.97	9.27	48.20
Vegetation, unwashed[3]						
60 ft.		3	**	9.20	**	108.07
140 ft.		3	**	10.03	132.00‡	73.73
220 ft.		3	**	9.03	**	53.40
Vegetation, washed[3]						
60 ft.		3	.90‡	6.07	21.23	56.33
140 ft.		3	**	7.07	22.67	49.13
220 ft.		3	.55‡	5.60	14.17	38.83
Dustfall 60 ft.[4]		3	.0386‡	.1682	2.7708	1.6625
Dustfall 140 ft.		3	**	.3111	1.6883	1.0660
Dustfall 220 ft.		3	**	.1156	1.9713	1.1115

[1] Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2] Mean value of all blood and milk samples in ug/100 ml; Jan. 5, 1972.

[3] Mean value of hair, soil, root and vegetation samples as ug/g; Jan 5. 1972.

[4] Mean value as $mg/m^2/mo$; Jan. 4 - April 2, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

‡Two sample values were below lower detectable limit; therefore, the value presented is based on one sample.

Table 5. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Test Farm near Glover, Missouri, During April 1 - July 1, 1972.

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1]			
			Cd	Cu	Pb	Zn
Blood[2]	1	2	**	64.25	46.00	428.50
Blood	2	2	.50	64.25	87.00	525.00
Blood	3	2	.60	72.50	43.00	394.00
Blood	4	2	.70	52.90	59.00	353.50
Milk[2]	1	2	**	8.90	20.50	323.00
Milk	2	2	**	7.90	20.50	341.50
Milk	3	2	.30*	9.40	24.00	134.00
Milk	4	2	.30*	7.70	35.00	304.50
Hair[3]	1	2	2.00	6.20	84.00	145.00
Hair	2	2	2.00	7.50	93.00	133.50
Hair	3	2	4.50	8.45	143.00	126.00
Hair	4	2	2.70	5.60	66.00	117.50
Soil 60 ft.[3]		3	.65	14.57	99.00	41.23
Soil 140 ft.		3	.63	13.23	84.67	30.67
Soil 220 ft.		3	.83	10.47	82.20	27.57
Roots 60 ft.[3]		3	2.53	22.67	343.33	86.83
Roots 140 ft.		3	3.60	27.50	293.67	94.17
Roots 220 ft.		3	2.47	22.33	290.00	72.33
Vegetation, unwashed[3]						
60 ft.		3	**	21.35+	851.23	103.23
140 ft.		3	8.13	18.37	807.07	104.60
220 ft.		3	10.55+	16.10	810.73	100.80
Vegetation, washed[3]						
60 ft.		3	7.33	16.50	696.67	84.33
140 ft.		3	6.57	17.00	718.33	88.00
220 ft.		3	7.97	14.83	720.00	84.67
Dustfall 60 ft.[4]		3	.5954	2.4622	100.5931	12.9044
Dustfall 140 ft.		3	.5589+	2.2803+	84.0318+	14.0149+
Dustfall 220 ft.		3	.4825	1.9644	72.7931	10.7524

[1]Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2]Mean value of all blood and milk samples in ug/100 ml; Apr. 1, 1972.

[3]Mean value of hair, soil, root and vegetation samples as ug/g; Apr. 1, 1972.

[4]Mean value as $mg/m^2/mo$; April 1 - July 1, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

Table 6. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Control Farm near Ellington, Missouri, During April 2 - June 30, 1972.

Type Sample	Animal ID No.	No. of Samples	Mean Value[1] Cd	Cu	Pb	Zn
Blood[2]	1	2	.35	96.25	9.00	328.00
Blood	2	2	1.25	103.75	9.00	431.50
Blood	3	2	.80*	128.00	10.00	313.00
Blood	4	2	.80*	111.00	13.50	444.00
Milk[2]	1	2	.30*	10.65	7.00*	470.50
Milk	2	2	**	5.95	4.00	245.00
Milk	3	2	**	16.35	7.50	374.00
Milk	4	2	**	8.15	9.00	414.00
Hair[3]	1	2	.04	7.55	2.80	115.00
Hair	2	2	.05	5.80	2.00	96.50
Hair	3	2	.07	6.50	1.85	100.75
Hair	4	2	.06	7.40	1.85	95.25
Soil 60 ft.[3]		3	.57‡	7.33	19.20	21.57
Soil 140 ft.		3	**	8.67	19.10	22.43
Soil 220 ft.		3	**	7.47	18.30	27.67
Roots 60 ft.[3]		3	.71	16.33	17.67	41.00
Roots 140 ft.		3	.68	15.00	28.50	34.33
Roots 220 ft.		3	.70	15.80	20.83	39.17
Vegetation, unwashed[3]						
60 ft.		3	**	7.06‡	59.30‡	36.77
140 ft.		3	**	7.35+	**	30.30
220 ft.		3	**	7.50‡	**	30.10
Vegetation, washed[3]						
60 ft.		3	**	5.67	20.67	30.33
140 ft.		3	**	6.33	27.33	24.50
220 ft.		3	**	6.17	25.50	24.83
Dustfall 60 ft.[4]		3	.0594	.3464	4.1959	2.1217
Dustfall 140 ft.		3	.0483	.3958	2.1256	2.0267
Dustfall 220 ft.		3	.0507	.2573	2.3355	1.3973

[1]Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2]Mean value of all blood and milk samples in ug/100 ml; Apr. 2, 1972.

[3]Mean value of hair, soil, root and vegetation samples as ug/g; Apr. 2, 1972.

[4]Mean value as $mg/m^2/mo$; April 2 - June 30, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

‡Two sample values were below lower detectable limit; therefore, the value presented is based on one sample.

Table 7. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Test Farm near Glover, Missouri, July 1 - October 1, 1972

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1]			
			Cd	Cu	Pb	Zn
Blood[2]	1	2	1.85	73.00	33.50	425.00
Blood	2	2	.30*	86.00	30.50	455.00
Blood	3	2	.30	67.50	30.50	420.00
Blood	4	2	.35	78.00	18.00	310.00
Milk[2]	1	2	**	9.00*	3.50	390.00
Milk	2	2	**	**	3.00	335.00
Milk	3	2	**	**	20.00*	230.00
Milk	4	2	.20*	**	3.50	300.00
Hair[3]	1	2	.47	6.10	53.00	102.75
Hair	2	2	.84	9.30	69.00	93.35
Hair	3	2	.58	8.15	63.50	84.10
Hair	4	2	.79	8.40	78.50	93.00
Soil 60 ft.[3]		3	.89	13.80	157.17	42.17
Soil 140 ft.		3	.73	11.53	110.33	27.13
Soil 220 ft.		3	.87	11.43	117.33	30.43
Roots 60 ft.[3]		3	1.77	22.73	242.50	105.00
Roots 140 ft.		3	1.87	17.00	98.00	35.17
Roots 220 ft.		3	1.17	12.73	55.83	33.50
Vegetation, unwashed[3]						
60 ft.		3	1.03	7.27	130.97	47.73
140 ft.		3	.65‡	6.40+	132.20+	32.60+
220 ft.		3	1.34	5.70	95.97	26.83
Vegetation, washed[3]						
60 ft.		3	1.03	5.93	93.27	37.70
140 ft.		3	.65‡	5.90+	113.25+	29.50+
220 ft.		3	1.34	4.37	64.67	20.93
Dustfall 60 ft.[4]		3	1.6082	2.3932	121.9600	19.9501
Dustfall 140 ft.		3	1.3249	1.8572	89.0289	15.2785
Dustfall 220 ft.		3	1.2407	1.6542	78.4986	13.5171

[1] Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2] Mean value of all blood and milk samples in ug/100 ml; July 1, 1972.

[3] Mean value of hair, soil, root and vegetation samples as ug/g; July 1, 1972.

[4] Mean value as $mg/m^2/mo$; July 1 - Oct. 1, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

‡Two sample values were below lower detectable limit; therefore, the value presented is based on one sample.

Table 8. Results of Cadmium (Cd), Copper (Cu), Lead (Pb), and Zinc (Zn) Analyses of Samples Collected on Control Farm near Ellington, Missouri, July 1 - September 30, 1972.

Type of Sample	Animal ID No.	No. of Samples	Mean Value[1] Cd	Cu	Pb	Zn
Blood[2]	1	2	.30	72.00	6.00	380.00
Blood	2	2	.40*	126.00	8.00	355.00
Blood	3	2	.35	93.00	5.00	350.00
Blood	4	2	.40*	101.50	7.50	405.00
Milk[2]	1	2	.20	10.50	**	450.00
Milk	2	2	**	**	**	230.00
Milk	3	2	.50	12.50	**	335.00
Milk	4	2	.30*	**	**	360.00
Hair[3]	1	2	.06	7.75	1.25	83.70
Hair	2	2	.06	5.90	.55	78.25
Hair	3	2	.02*	7.60	.57	85.70
Hair	4	2	.02	8.40	1.15	82.70
Soil 60 ft.[3]		3	**	6.57	17.80	17.87
Soil 140 ft.		3	.37‡	7.20	15.57	16.57
Soil 220 ft.		3	**	5.73	14.43	12.63
Roots 60 ft.[3]		3	.53+	9.03	8.75+	84.57
Roots 140 ft.		3	.87	12.67	10.73	38.00
Roots 220 ft.		3	.69	13.17	11.50	41.25
Vegetation, unwashed[3]						
60 ft.		3	**	5.38	5.00‡	37.80
140 ft.		3	.70‡	4.40	5.40+	25.73
220 ft.		3	**	5.50	6.60+	29.93
Vegetation, washed[3]						
60 ft.		3	**	4.67	5.00‡	31.67
140 ft.		3	.70‡	4.40	5.40+	23.67
220 ft.		3	**	4.87	6.60+	21.87
Dustfall 60 ft.[4]		3	.0322+	.2527	2.0869	1.9414
Dustfall 140 ft.		3	**	.3102	1.3326	1.4283
Dustfall 220 ft.		3	**	.1915	1.7423	1.2981

[1]Calculated on a dry weight basis, except for blood and milk values which are on a wet weight basis.

[2]Mean value of all blood and milk samples in ug/100 ml; July 1, 1972.

[3]Mean value of hair, soil, root and vegetation samples as ug/g; July 1, 1972.

[4]Mean value as $mg/m^2/mo$; July 1 - Sept. 30, 1972.

*One sample value was below lower detectable limit; therefore, the value presented is based on one sample.

**All sample values were below lower detectable limits.

+One sample value was below lower detectable limit; therefore, the value presented is based on two samples.

‡Two sample values were below lower detectable limit; therefore, the value presented is based on one sample.