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Ecological Research Series

EFFECTS OF WASTEWATER AND COOLING WATER CHLORINATION ON AQUATIC LIFE

Environmental Research Laboratory
Office of Research and Development
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EFFECTS OF WASTEWATER AND COOLING WATER CHLORINATION ON AQUATIC LIFE

by

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FOREWORD

Our nation's freshwaters are vital for all animals and plants, yet our diverse uses of water---for recreation, food, energy, transportation, and industry---physically and chemically alter lakes, rivers, and streams. Such alterations threaten terrestrial organisms, as well as those living in water. The Environmental Research Laboratory in Duluth, Minnesota develops methods, conducts laboratory and field studies, and extrapolates research findings

--to determine how physical and chemical pollution affects aquatic life

--to assess the effects of ecosystems on pollutants

--to predict effects of pollutants on large lakes through use of models

--to measure bioaccumulation of pollutants in aquatic organisms that are consumed by other animals, including man

This report summarizes the recent literature on the effects of total residual chlorine on aquatic life and should be useful to people with interests in aquatic toxicology and the control and regulation of water pollution.

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CONTENTS

ACKNOWLEDGMENTS

This review, like any that attempts to be current, is greatly dependent upon researchers and other interested parties who have permitted the citation of their unpublished data and conclusions. There are many, as the following references indicate. These people are to be commended for their concern for this environmental problem, and I wish to thank them sincerely for their cooperation.

SECTION I

INTRODUCTION

In the past few years there has been a great increase in the amount of field and laboratory research on the effects of wastewater and cooling water chlorination on aquatic life. The majority of this research has been on the effects of intermittent chlorination used in the control of biofouling organisms. Some data for intermittent chlorination can be applicable to wastewater chlorination if they describe relative sensitivity of various aquatic organisms or life stages of single species. It is probably valid to assume that these sensitivity relationships would be comparable for aquatic life under conditions of continuous exposure, the characteristic of chlorinated municipal wastewaters.

The increased concern about chlorine toxicity results from three principal causes: (a) field observations demonstrating significant areas of biological degradation; (b) the erroneous impression that total residual chlorine (TRC) is very rapidly converted to chlorides; and (c) the increasing reliance on discharge-permit limitations in the National Pollutant Discharge Elimination System of the U.S. Environmental Protection Agency. Discharge permits have been influential in controlling the discharge of TRC from power-generating stations.

Until recently it was assumed that TRC would not persist in most environments because of the chlorine demand of the receiving water, which would reduce TRC to chloride, a relatively nontoxic anion. Studies have described (1, 2) the rate of decay of TRC by both dilution and chlorine demand of the receiving stream. On-site studies at several municipal waste treatment plants were conducted with plant effluents and stream water. Concurrent effects of chlorine demand of the receiving water, dilution, and time were studied. Dilution ratios of effluent to receiving water were 1:1, 1:4, and 1:9. One significant observation of these studies was that TRC persisted in the diluted wastewater throughout the 3-hr period. Also noteworthy was the almost complete lack of chlorine demand of the receiving water; the major effect on chlorine decay rate was from dilution only.

An additional problem in wastewater disinfection is that little effort has been made to optimize the process in existing plants (3). Personnel engaged in wastewater treatment have generally attempted to meet increasingly stringent bacteriological requirements of regulatory agencies by increasing the chlorine feed rate. Other changes such as mixing and contact time would require structural modifications. The need to optimize the disinfection process so that the toxic effect on aquatic communities is reduced to a minimum is obvious. Rising costs for disinfection procedures such as dechlorination may result in greater efforts toward optimization. Increasing chlorine feed rates to meet bacteriological limitations has resulted in maximum TRC values of 5.17 mg/l in central Illinois (4), of over 10 mg/l in southern Wisconsin (5), and as high as 15 mg/l in wastewater effluent in California (6).

The degree of environmental damage caused by chlorine has passed relatively unnoticed for other reasons. Harr (7) concluded that "chlorination is the most universally accepted method for the disinfection of wastewater." When a process has been as effective in protecting public health as chlorination, many data are needed to convince managers that it needs reappraisal.

There is an interesting historical note with regard to the regulation of chlorine discharges. In 1868, the Rivers Pollution Commission was appointed in the United Kingdom to inquire into "the best means of preventing the pollution of rivers." A report and conclusions were submitted in 1870 (8) with recommendations for the Mersey River and Ribble River basins. The Commission concluded that "it will be necessary to prescribe definite standards of purity below which no liquid shall be admissible into any river or stream." These standards of purity "have been framed with a due regard to the extent to which the cleansing of foul liquids can be effected without the imposition of undue restrictions upon the manufacturer." Numerical concentration limits were suggested for a wide variety of materials and conditions. If these concentration limits and conditions were exceeded the liquids would be "deemed polluting and inadmissible into any stream." For free chlorine the limit for any liquid, after acidification was 10 mg/l. The Commission had sufficient insight to believe that "as science progresses improved methods of purifying polluting liquids will be discovered, and that eventually standards of purity considerably higher....may, if necessary, be enforced."

The following discussion of the biological effects of chlorine is a continuation of an earlier review by the author (9). The reader is referred to that paper for publications before 1973. The present review is divided into various sections to permit those readers with only specific interests in chlorination to find those topics with a minimum of effort. These sections are: review papers, chlorinated municipal wastewaters, continuously chlorinated water, intermittently chlorinated water, dechlorination, avoidance, formation of chlorinated organic compounds, miscellaneous, aquatic life criteria and application factors, and regulations.

SECTION II

SUMMARY

The literature since 1972 pertaining to wastewater and cooling water chlorination is discussed under the following headings: review papers, chlorinated municipal wastewaters, continuously chlorinated water, intermittently chlorinated water, dechlorination, avoidance, formation of chlorinated organic compounds, aquatic life criteria and application factors, and regulations.

Field and laboratory research results support a single criterion of 0.003 mg/l for continuous exposure of freshwater organisms. The former distinction between warmwater and coldwater systems is no longer appropriate; recent data indicate that several freshwater fish species are as sensitive as trout and salmon.

The present concern for the formation of chlorinated organics in water and wastewaters is justifiable and the greatest present need is to determine the ecological significance, if any, of these results. The future course of wastewater chlorination will be greatly influenced by the recent proposed changes in the Environmental Protection Agency's regulations on secondary treatment. The changes intend that disinfection only be considered when public health hazards need to be controlled, and that the exclusive use of chlorine should not be considered where protection of aquatic life is of primary consideration. Where these uses co-exist, alternate means of disinfection must be considered.

SECTION III

CONCLUSIONS

1. Total residual chlorine (TRC) is acutely lethal to some aquatic organisms at concentrations below the usual detectable analytical limit.

2. The toxicity of TRC in seawater is less clear than in freshwater because of the unclear picture of the chemistry of halogens in seawater.

3. The acute toxicity of TRC in clean water and chlorinated domestic wastewaters is generally the same. This appears to be true of chronic toxicity also.

4. Dechlorination appears to eliminate the acute and chronic toxicity of TRC to aquatic life.

5. Intermittent chlorination for biofouling control in freshwater can generally be significantly reduced with no loss of control.

6. Several species of freshwater minnows are as sensitive as trout and salmon to lethal TRC concentrations.

7. Most freshwater invertebrates are no more sensitive to TRC than freshwater fish.

8. Avoidance of intermittent chlorination in the laboratory and continuous wastewater chlorination in the field has been demonstrated. The implications of the former have not been demonstrated.

9. Chlorination of water and wastewater results in the formation of chlorinated compounds that have been shown to be bioconcentrated by freshwater fish.

SECTION IV

RECOMMENDATIONS

1. Several important general research needs are evident from this review:

   a) Halogen chemistry in seawater.

   b) Chemical and toxicological data on alternatives to disinfection by chlorination.

   c) More sensitive and specific analytical methods for disinfectants.

   d) Exposure conditions necessary to control various species of biofouling organisms.

2. When disinfection and the protection of aquatic life are both necessary, the most feasible alternative at this time would be dechlorination.

3. The single criterion for continuous exposure of freshwater organisms to TRC should be 0.003 mg/l.

4. The most appropriate criteria for intermittent exposure of aquatic organisms to TRC are time dependent.

SECTION V

REVIEW OF THE LITERATURE

REVIEW PAPERS

Tsai (10) reviewed the literature on the effects of sewage treatment plant effluents on fish. He discussed not only the toxic effects of TRC, but also the toxic effects of ammonia, synthetic detergents, hydrogen sulfide, sewage sludge, and dissolved oxygen. Additional topics of this review included fish culture in wastewater effluents and toxic flagellates. One of his more important observations about chlorine toxicity was that very few studies have been conducted on the effects of continuous exposure of TRC on estuarine and marine fishes. This situation continues to persist; most of the present marine research, to be discussed later, is oriented to the intermittent effects of power plant chlorination or is conducted under laboratory conditions with clean dilution water rather than with chlorinated wastewater effluent itself.

The literature on the use of calcium hypochlorite in fisheries was summarized by Podoliak (11). His title and discussion suggest a very narrow topic, but his listing of approximately 1,230 citations would be useful to any generalist or specialist involved in manufacturing, use, effects, or regulation of chlorine. Chlorine was used to eradicate unwanted fish 40 years ago, but its principal use in fisheries today is to control or eliminate infectious diseases and parasites in hatcheries.

Becker and Thatcher (12) reviewed and tabulated data on the toxicity to aquatic life of a wide variety of chemical additives, including chlorine and bromine, that may be used in nuclear power plants. In tabulating the toxicity data, they described the chemical compound, test organism, test conditions, concentrations, and the type of test conducted.

Whitehouse (13) prepared a review of the literature on chlorination of cooling water and presented data on the effects of free and combined chlorine on a wide variety of aquatic organisms. He concluded that marine and freshwater species are apparently equally vulnerable to the action of chlorine and that smaller organisms are affected in shorter exposure times than larger ones. He concluded that all aquatic organisms are susceptible to chlorine and that chlorine apparently has a common mode of action on living material, although certain organisms with specialized tissues such as gills or byssus glands may not fit this generalization.

An annotated bibliography was prepared by Mattice and Pfuderer (14) that covers the chemistry and effects of chlorine in aquatic systems. This bibliography contains abstracts of 190 papers and reports written during the past 50 years. Evins (15) surveyed and summarized published data on the toxicity of chlorine and chloramines to some freshwater organisms, excluding bacteria and fungi. Davis and Middaugh (16) reviewed the use of chlorine as a

disinfectant and antifouling chemical in marine ecosystems. They discussed briefly the chemistry of chlorine in marine waters with emphasis on the production of bromine and bromamines by the chlorination of seawater. They summarized the toxicity of TRC for phytoplankton, invertebrates, and estuarine fish. Brooks and Seegert (17) discussed the various factors such as chemical forms, time of exposure, and temperature that are important in determining the toxicity of TRC to aquatic life. They presented a synthesized view of the toxicity of TRC as it relates to clean waters and sewage effluents. The principal differences in toxicity were due to differences in chlorination, whether intermittent or continuous, and in experimental temperatures, which covered a wide test range.

The Atomic Safety and Licensing Board requested a critical review (18) of current literature and research on the lethal, sublethal, and behavioral effects of chlorine and other biocides on aquatic life and on the chemistry and biochemistry of the products formed following release and degradation of chlorine in the aquatic environment. The principal purpose of this review was to evaluate biofouling control for power-generating stations on the Columbia River.

CHLORINATED MUNICIPAL WASTEWATERS

Servizi and Martens (19) placed rainbow trout at various points downstream from three sites where chlorinated domestic wastewater effluent was being discharged. At one site the discharge from a retention lagoon essentially resulted in a dechlorinated final effluent that was nontoxic to the rainbow trout. The test fish died downstream from the other two sites that had no retention ponds. During one test period at one of the latter sites the chlorinator was not operating, and no fish died. They concluded, in general, that lethality was common at stations where TRC concentrations were 0.02 mg/l or greater.

One hundred forty-nine domestic wastewater treatment plants were studied in Virginia, Pennsylvania, and Maryland (20). Fish-community diversity and fish occurrence were related to observed TRC concentrations. In most of the plants studied a TRC concentration of 0.5 to 2.0 mg/l was maintained in the effluent. In streams receiving chlorinated effluent no living organisms were observed in the immediate vicinity of the outfall. No fish were found in water at TRC concentrations at or above 0.37 mg/l. The species diversity index fell to zero at 0.25 mg/l. An analysis of Tsai's graphically presented data indicated that the brown and brook trout did not occur at TRC concentrations above approximately 0.02 mg/l. Ten fish species, including five minnows, a bullhead, two darters, and the two trout were not observed at concentrations above 0.05 mg/l. At 10 facilities with chlorination and an effluent-holding lagoon that resulted in dechlorination, fish-species diversity did not differ above and below the outfalls.

Olson (21) studied the effect of chlorinated wastes from the Marshalltown (Iowa) water pollution control plant on native and caged fish in the Iowa River. The dilution of the effluent during these studies ranged from 25:1 to 282:1. The observations were made just as the plant began a chlorination program, and the TRC concentrations determined amperometrically varied from 0.00 to 5.0

mg/l. No adverse effects on native fish were observed, and caged channel catfish did not survive within a distance of 150 ft after 96 hr of exposure.

Norris et al. (22) discussed ecological investigations of municipal waste disposal in San Francisco Bay and the Gulf of the Farallones. Static bioassays were conducted with a variety of marine and estuarine fish and invertebrate species. Continuous-flow bioassays were conducted on Dungeness crab zoea and three-spined stickleback. Unfortunately, all of the toxicity results are expressed as percentage of effluent, and no mention is made of residual chlorine concentrations.

During 1973 and 1974 massive fish kills involving bluefish, sea trout, croaker, menhaden, and others were observed in the James River estuary of Virginia (23). Extensive field studies determined that the cause was chlorinated wastewater discharged to the system by two sewage plants. Field bioassays were conducted with bluegill sunfish, and unchlorinated and chlorinated effluents in the plant confirmed the lethality of the discharge. When the chlorination feed was reduced, the fish kill was almost completely eliminated during the next 24 hr. This observation was made during both years. Laboratory bioassays with a variety of estuarine species provided 48- and 96-hr LC50 values between 0.005 mg/l for oyster and clam larvae and 0.28 mg/l for pipefish. Details of these studies will be discussed later in this review.

Unchlorinated sewage was a relatively weak inhibitor of fertilization of gametes of the sea urchin, Strongylocentrotus purpuratus (24). Chlorinated sewage was more detrimental, and adverse effects were observed at TRC concentrations as low as 0.05 mg/l. The principal effect was on sperm. Fertilization was also inhibited at 0.20 mg/l for Urechis caupo and Phragmatopoma californica. All exposures were for only 5 min.

Nominal TRC concentrations of 0.02, 0.1, and 3.0 mg/l for a 1-hr contact time were evaluated for disinfection capability with primary sewage (25). An acceptable MPN (most probable number) was obtained only at 3.0 mg/l, which nominal concentration was highly toxic to sockeye salmon. Measured TRC concentrations varied considerably and ranged from 0.06 to 2.05 mg/l. Golden shiners were exposed to chlorinated primary and secondary effluent by Esvelt et al. (26), and continuous-flow, 96-hr LC50 values averaged 0.19 mg/l with a standard deviation of 0.08 mg/l.

Stone et al. (27) reported on studies that focused on the long-term effects resulting from the discharge of various municipal effluents into the waters of Central San Francisco Bay. Central Bay model aufwuchs (represented by decomposers, producers, and herbivores) were tested with unchlorinated and chlorinated secondary effluent. Above 4 percent effluent the unchlorinated waste stimulated growth of aufwuchs biomass. The average TRC concentration was 0.06 mg/l in the bay model tanks receiving 1 percent effluent; this concentration reduced biomass accumulation to less than 30 percent of that measured in the controls. A reduction in chlorophyll a concentration to about 50 percent of the control was also observed in a 1-percent effluent with a TRC concentration of 0.06 mg/l. No aufwuchs biomass was found in tanks receiving higher TRC concentrations. Golden shiners were also studied; tap water was used for dilution of the chlorinated primary effluent. Ninety-six-hr LC50 values ranged from 0.16 to 0.26 mg/l with a mean of 0.21 mg/l.

Chronic, life-cycle toxicity tests with the fathead minnow, a cladoceran (Daphnia magna), and an amphipod and numerous acute tests with fish and invertebrate species were conducted by Arthur et al. (28). Non-disinfected, chlorinated, dechlorinated, and ozonated secondary effluent was used. The lowest mean TRC concentrations having a measurable adverse chronic effect were 0.042 mg/l for the fathead minnow, 0.019 mg/l for the amphipod, and approximately 0.010 mg/l for Daphnia magna. The highest mean TRC concentrations having no measurable effect were 0.014 mg/l for the fathead minnow, 0.012 mg/l for the amphipod, and 0.002 to 0.004 mg/l for Daphnia magna. This corresponds to a concentration range of 1.2 to 2.5 percent chlorinated effluent. One-hr to 7-day LC50 values were determined for the brook trout, coho salmon, fathead minnow, white sucker, walleye, yellow perch, largemouth bass, amphipod, stonefly, caddisfly, crayfish, and snails. The 7-day LC50 values for the fish ranged from 0.082 to 0.261 mg/l. The invertebrates were generally more resistant with 7-day LC50 values from 0.21 to >0.81 mg/l.

Ward et al. (29) and Ward (30) discussed a set of toxicity tests nearly comparable to those of Arthur et al. (28) at the Grandville (Michigan) Wastewater Treatment Plant, an activated sludge facility receiving wastewater almost totally derived from domestic sources. The results of their chronic test with the fathead minnow were consistent with those of Arthur et al. (28). The lowest concentration of TRC that caused an adverse effect was 0.045 mg/l, at which concentration larval growth and survival were affected. A concentration of 0.01 mg/l was considered safe. Acute toxicity tests with the fathead minnow, lake trout, goldfish, rainbow trout, coho salmon, largemouth bass, crappie, an unidentified sunfish, walleye, pugnose shiner, common shiner, and golden shiner also yielded comparable results to those of Arthur et al. (28), with 96-hr LC50 values between 0.040 and 0.278 mg/l. The LC50 values for the four species of minnows were as low as or lower than those for the trout and salmon, which would indicate that these warmwater fish species are as sensitive as the trout and salmon. The most resistant fish were the sunfish and largemouth bass. Chlorobrominated effluent had no chronic effect on fathead minnows at concentrations of residual bromine at or below 0.043 mg/l; 96-hr LC50 values for 14 fish species were from 0.047 to 0.283 mg/l residual bromine. Fathead minnows exposed to sublethal concentrations of chlorine and bromine before LC50 tests were more resistant to lethal concentrations than those fish not previously exposed. Similar acute and chronic toxicity tests to those described above have begun at the Wyoming (Michigan) Wastewater Treatment Plant; the influent to this plant is about equally derived from industrial and municipal operations.

Primary, secondary, and chlorinated secondary effluents from a municipal sewage treatment plant were tested to determine their impact on off-flavor in rainbow trout as determined by the Sensory Evaluation Section of the Department of Food Science and Technology, Oregon State University (31). Only chlorinated secondary effluent concentrations of 20 percent by volume and above produced tainted fish. In one experiment no flavor impairment occurred at 33 percent by volume, and all fish died at higher concentrations. The addition of chlorine to the secondary effluent appeared to reduce the impairment since concentrations of about 15 percent by volume of unchlorinated primary and secondary effluent produced an off-flavor.

CONTINUOUSLY CHLORINATED WATER

Eren and Langer (32) studied the effects of chlorinated water on a mixed population of two species of tilapia. Fish kills involving these species had

been observed in the operational reservoirs of the Israel National Water System. An especially severe kill occurred in the Tsalmon Reservoir, and chlorine was suspected since the channel leading to this reservoir had been previously chlorinated with measured TRC concentrations up to 0.3 mg/l. Various laboratory exposures for 4 to 18 hr were conducted at different temperatures and ratios of free and combined chlorine. The authors concluded that smaller fish were more sensitive, that fish were more sensitive at the higher test temperatures, and that free chlorine was more toxic than combined chlorine.

When a concentration of 0.4 mg/l TRC was maintained continuously to eliminate fouling organisms at a marine power station, productivity of entrained marine phytoplankton decreased 83 percent (33). Productivity was measured at six other continuously applied TRC concentrations, and the lowest concentration tested, which was below the authors' detectable limit of 0.1 mg/l, caused a decrease of 79 percent in productivity. When chlorine was not applied during the study period, phytoplankton productivity was essentially unaffected.

Gentile et al. (34) discussed the results of field and laboratory investigations on chlorine toxicity to marine organisms related to power plant condenser cleaning. In the laboratory growth rate of 11 species of phytoplankton decreased 50 percent during a 24-hr exposure to TRC concentrations between 0.075 and 0.330 mg/l. Phytoplankton were also studied at several power-generating stations. Median survival times for three species of estuarine copepods ranged from 120 to 360 min at 1.0 mg/l to 0.7 to 5 min at a TRC concentration of 10 mg/l. Two species of ichthyoplankton (winter and yellowtail flounder) were studied under laboratory conditions. The 24-hr LC50 values for the yellowtail flounder were 0.2 and 0.1 mg/l. Attempts were made to evaluate these effects in the field.

The combined effects of chlorine and temperature on rainbow and brook trout were studied by Wolf et al. (35). Sensitivity of brook trout decreased as size of the fish increased. Ninety-six-hr LC50 values as determined in continuous-exposure tests were between approximately 0.04 and 0.065 mg/l with a mean of about 0.05. Thatcher et al. (36) acclimated brook trout to 10, 15, and 20° C and tested these fish at 7, 10, 15, and 20° C. The TRC 96-hr LC50 values from 13 continuous-flow toxicity tests run at 10 and 15° C were not significantly different regardless of the previous acclimation temperature. At 20° C the brook trout were more sensitive. The mean LC50 values at 10 and 15° C were about 0.15 mg/l; the mean value at 20° C was about 0.10 mg/l. Schneider et al. (37) cited these same results, but included data from some interesting studies with crayfish. The 96-hr LC50 value for TRC was 0.96 mg/l and indicated that the crayfish were very resistant. However, many crayfish died during the 6-wk period after the exposure ended. When the LC50 was recalculated to take these deaths into account, the value became 0.25 mg/l.

Warren (38) exposed several fish species and crayfish to chloramines and determined acute and subacute effects on growth and survival. Alkalinity and pH were varied, but over the experimental range (pH, 7.5 to 8.1; alkalinity, 140 to 320 mg/l) the 96-hr LC50 values for cutthroat trout and coho salmon did not vary significantly (0.078 to 0.089 mg/l). Ninety-six-hr LC50 values for brook trout ranging in age between 8 and 60 weeks were between 0.082 and 0.091 mg/l, which indicated no influence of age on sensitivity, a result different from that of Wolf et al. (35). Growth, food consumption, or food-conversion efficiency at different feeding levels of coho salmon juveniles were not affected by exposure to TRC concentrations of 0.005 and 0.010 mg/l. Fish exposed to 0.020 mg/l chloramines had significantly lower growth, food consumption, and

food-conversion efficiency. Residual chlorine had no apparent effect on the fertilization process (artificial), embryo survival, time of hatching, or alevin mortality at TRC concentrations (only during fertilization) as high as 0.394 mg/l. Results of partial chronic tests with crayfish indicated mortality at a chloramine concentration as low as 0.100 mg/l and at 0.050 mg/l egg clusters carried by female crayfish contained few, if any, live embryos. The control and 0.025 mg/l exposures showed no effects on the embryos. Additional studies on laboratory streams are continuing.

Blacknose dace were exposed (39) to both free chlorine and chloramine concentrations for several periods of time to determine survival times after short exposure and subsequent return to chlorine-free water and lethal exposure times. The need for determining both types of results became apparent when the authors observed dead fish among the fish returned to clean water after brief exposures to chlorine. At higher test concentrations free chlorine was more toxic than chloramines, but it was less toxic at lower concentrations.

Bluegill and channel catfish were exposed to chloramines under continuous-flow conditions by Roseboom and Richey (40). Different temperatures and fish sizes were compared. The 96-hr LC50 values for the more resistant bluegill ranged from 0.18 to 0.33 mg/l. The authors indicated that the smaller bluegills were more sensitive and that chloramine was more toxic at a higher temperature. The estimated 96-hr LC50 for the channel catfish was about 0.09 mg/l, and temperature did not appear to influence the sensitivity of this fish species.

Carlson (41) exposed mature threespine sticklebacks to chloramine concentrations of 0.001, 0.0043, and 0.0114 mg/l for 3.5 months to observe their reproductive behavior under laboratory conditions simulating their natural environment. Isopod and amphipod populations were included in the test chambers as a partial food source for the sticklebacks. By the end of the test no differences in reproductive behavior of the fish could be observed at any of the measured chloramine concentrations. In addition, no differences were detectable in the standing crops of amphipods (Crangonyx sp.) or isopods between the control and experimental chambers. Early in the study the growth of periphyton on the chamber walls was delayed at the tested concentrations as compared to the controls. Estimates of periphyton standing crop at the end of the test reflected a trend of decreasing biomass with increasing chloramine concentrations; effects were observed at all tested concentrations.

Eggs and larvae of plaice, a flatfish, were continuously exposed to TRC concentrations at test temperatures of 5.4 to 9.3° C (42). The 3-day and 8-day LC50 values for eggs were 0.7 and 0.12 mg/l, respectively. Ninety-six-hr LC50 values for the larvae were 0.024, 0.028, and 0.034 mg/l. The egg membrane appeared to give considerable protection to the embryo, allowing development to continue normally over long periods, even in TRC concentrations that would be rapidly lethal to the hatched larvae. Hughes (43) used reconstituted water and acute, static bioassay methods to determine the sensitivity of striped bass larvae and 1-month-old striped bass fingerlings to 30 chemicals including chlorine. Nearly all the observed deaths due to chlorine occurred within 24 hr; the 96-hr LC50 values for larvae and fingerlings were 0.5 and 0.25 mg/l.

The toxicity of TRC to juvenile spot in flowing seawater was studied by Middaugh et al. (44). Incipient lethal concentrations were determined, and avoidance tests were conducted. Histopathological changes and the combined effect of thermal stress were also determined. After continuous exposure for up to 8 days, the incipient LC50 values for TRC were 0.12 mg/l at 10° C and 0.06 mg/l at 15° C. Pseudobranch and gill damage was indicated at a measured TRC concentration of 1.57 mg/l; no consistent tissue damage was detectable after exposure to 0.02 to 0.62 mg/l. Avoidance also appeared to be temperature dependent with avoidance to a TRC concentration of 0.18 mg/l at 10° C. At 15 and 20° C the spot avoided concentrations as low as 0.05 mg/l. After exposure to 15, 20, 25, and 28° C for 7.5, 15, 30, and 60 min, a significant increase in sensitivity was detectable related to increased temperature and exposure time.

Gregg (45) described studies with a variety of aquatic invertebrate species exposed continuously and intermittently at different temperatures to TRC. He also conducted heat-shock and chlorine-plus-heat-shock studies to simulate conditions to which organisms entrained in cooling waters are exposed. Median survival times and concentrations were determined; carbon-filtered tap water was used for dilution. Temperature exerted an influence on chlorine toxicity, although the degree of effect varied with different species and for some no temperature effect was detected. Mayflies, stoneflies, sowbugs, amphipods, caddisflies, water pennies (beetle), and snails were tested. The LC50 values for continuous exposures were intermediate between the LC50 values for intermittent exposures based on maximum or peak concentrations and mean concentrations. Extreme differences in chlorine sensitivity were found among the species tested, even among the mayfly species. The majority of the 4- and 7-day LC50 values for continuous exposure to TRC were between 0.010 and 0.10 mg/l. Since some of these tests were conducted at nearly lethal temperatures, some instances of high control mortality may have been due to heat.

The rotifer, Keratella cochlearis, was continuously exposed by Grossnickle (46) to concentrations of TRC for periods of 1, 4, and 24 hr. Any longer exposure would have been over 10 percent of the average life span of this species. Only the most resistant individuals could survive concentrations of 0.055 mg/l or above at any of the exposure times; control mortality was 0 to 12 percent. The 1-, 4-, and 24-hr LC50 values were 0.032, 0.027, and 0.0135 mg/l, respectively.

Several invertebrate estuarine species were exposed to various TRC concentrations by Roberts et al. (47) under continuous-flow, continuous-exposure conditions for up to 96 hr. The 48-hr LC50 for the copepod, Acartia tonsa, was less than 0.05 mg/l, the minimum detectable TRC concentration in this study. Larvae of the clam, Mercenaria mercenaria, and oyster, Crassostrea virginica, demonstrated 48-hr LC50 values of less than 0.005 mg/l. The 96-hr LC50 for the glass shrimp, Palaemonetes pugio, was 0.22 mg/l. Of the three fish species tested, the pipefish was the most tolerant with a 96-hr LC50 of 0.28 mg/l. The 96-hr LC50 values for silversides and naked gobies were 0.037 and 0.08 mg/l, respectively. The authors used amperometric titration and could not detect monochloramine; they assumed that only free chlorine was present in all experiments. Bender et al. (48) also discussed these results on estuarine fish and invertebrates, but included a brief discussion of the uptake of carbon by four species of algae. Nannochloris occulatus, Tetraselmia suecica, Pseudoisochrysis paradoxa, and Pyramimonas virginica in unialgal cultures and natural mixed populations were exposed to various TRC concentrations. The algae in monospecific cultures were tested at three

salinities and three temperatures in pasteurized river water. Carbon uptake rates were measured with a 4-hr incubation period. More details of the methods have been discussed (49). Results expressed in relation to the control values and EC50 values ranged from approximately 0.01 to 0.47 mg/l depending upon species, temperature, and salinity.

Gibson et al. (50) conducted preliminary toxicity tests and growth rate experiments with the coon stripe shrimp, Pandalus danae, to determine LC50 values for temperature, chlorine, and copper, and the sublethal effects of these factors on growth rate. Shrimp were acclimated at 8 and 15° C and exposed for 96 hr to TRC at test temperatures of 10, 15, and 20° C. The shrimp were most resistant when acclimated and exposed at the lowest test temperature; the 96-hr LC50 values were 0.26 to 0.36 mg/l. They were less resistant when acclimated at the low temperature and tested at the two higher temperatures; the 96-hr LC50 values were between approximately 0.11 and 0.19 mg/l. Intermediate results, 96-hr LC50 values between 0.15 to 0.21 mg/l, were obtained for those shrimp acclimated to and tested at 15° C, which was near the shrimps' optimum short-term growth temperature of 16° C. A TRC concentration of 0.18 mg/l was lethal to young shrimp at 16° C, and 0.08 mg/l reduced their growth over a 1-month period.

Beeton et al. (51) exposed freshwater copepods and rotifers to continuous concentrations of TRC. The copepod Cyclops bicuspidatus thomasi was quite sensitive with a 96-hr LC50 of 0.084 mg/l as monochloramine. The 96-hr LC50 for this same species was 0.069 mg/l when the TRC was a mixture of hypochlorite and monochloramine. The 4-hr LC50 values for the rotifer Keratella cochlearis was 0.019 mg/l.

Sodium hypochlorite was added by Sanders et al. (52) to hatchery water in experiments to evaluate its ability to control the infectious protozoan, Ceratomyxa shasta. Chlorination and filtration apparently inactivated infectious organisms when TRC ranged from 2.2 to 5.3 mg/l. Granular activated carbon was used to dechlorinate the hatchery water after a 60-min contact time.

The effect of free chlorine and chloramine on the uptake of nitrate and ammonia by freshwater phytoplankton was studied by Toetz et al. (53). Free chlorine and chloramine were added to bottled lake water in concentrations that ranged from 0.01 to 0.1 mg/l. These bottles were incubated for about 24 hr after which uptake was determined. These single doses of chlorine reduced nitrate uptake by phytoplankton at a concentration as low as 0.028 mg/l.

INTERMITTENTLY CHLORINATED WATER

The following studies evaluating the impact of intermittent chlorination on freshwater and marine organisms are most applicable to cooling waters. As discussed earlier, however, some of the data are useful also for the evaluation of chlorinated municipal wastewaters. Any adverse concentrations observed would obviously be adverse under continuous-exposure conditions. The relative sensitivity of species, or life stages of a species, intermittently exposed may be the same under continuous-exposure conditions, that is, a species very sensitive, or very resistant, under the intermittent-exposure conditions could be expected to respond similarly to conditions comparable to those found in chlorinated wastewaters.

Chlorination of cooling water to retard growth of fouling organisms was first tried in the United States in 1924. All but about 10 percent of the electric generating plants chlorinate on a programmed basis. Exceptions are those plants using cooling water that contains sufficient suspended solids to physically scour the condenser tubes. The total amount of chlorine used for this purpose is currently about 100,000 tons per year (54).

Cole (55) summarized chlorination practices used to control biofouling in freshwater and seawater systems and under once-through and recirculating conditions. He believed that there is a tendency for operators to overchlorinate because of the difficulty in determining the required chlorination schedule. The author concluded that it appeared reasonable to expect that heat exchangers can be maintained in a clean condition using less chlorine than has been used in the past, since minimization programs have resulted in reductions in the duration of chlorination periods, frequency of chlorination, and chlorine feed rates. Draley (56) discussed biofouling control in cooling towers and closed cycle systems in general and specifically at a nuclear power station with a mechanical draft cooling tower and a coal-fired plant with a natural draft cooling tower. He was concerned principally in describing the buildup and decay of chlorine in these systems. Monitoring of residual chlorine in effluents for compliance with discharge regulations and the necessity of using continuous recorders was discussed by Baker (57). The levels at which these analyzers are required to operate approach the precision and sensitivity of the methods used to calibrate them. Practical problems associated with operation and maintenance of continuous recorders under normal municipal-industrial conditions would, at present, limit the lowest practical level that can be recorded with assurance to about 0.1 mg/l.

Mattice and Zittel (58) critically summarized the experimental and analytical methodology used to develop the data in the literature on chlorine toxicity to aquatic organisms. Available data for freshwater and marine organisms were summarized, and acute and chronic toxicity thresholds were approximated that would result in no toxicity. Log TRC concentrations versus log time plots demonstrated different acute and chronic thresholds for freshwater and marine species. The chronic toxicity threshold for fresh water was 0.0015 mg/l and that for salt water was 0.02 mg/l. The freshwater organisms appeared to be more sensitive to chronic exposure, but marine organisms appeared more susceptible to acute doses of TRC. The authors used these thresholds to develop a site-specific procedure for evaluating chlorination schemes at power plants. An analysis of a hypothetical marine-based power plant was presented to demonstrate the procedural steps involved. The analysis was based on comparison of the concentrations and exposure times expected at this hypothetical plant and the toxicity threshold developed for marine organisms. Additional examples of a hypothetical marine power plant have also been discussed in a summary of the above predictive site-specific analysis by Mattice (59) in which he compared a single plant with three possible dilution rates.

Brungs (60) reviewed the literature relating to condenser antifouling and discussed the possible additive effect of other stresses such as supersaturation, temperature, diseases, and parasites. This review included a brief discussion of needed research on antifouling control and its effects.

Dickson *et al.* (61) conducted an *in situ* toxicity test in the Clinch River (Tennessee) to evaluate the effects of intermittently chlorinated cooling tower blowdown. Bluegills and snails were placed in cages at various

distances downstream from the plant that was treating the cooling towers four times a day at 0.75 mg/l for 30 min. The free chlorine concentration ranged from 0 to 88 percent (0 to 0.35 mg/l) of the TRC at the point of discharge; the mean was 17.3 percent (0.08 mg/l). The highest observed free chlorine and TRC concentrations in the river were 0.07 and 0.35 mg/l, respectively. No bluegill deaths could be attributed to chlorine, but the blowdown was lethal to 50 percent of the snails in 72 hr when exposed to TRC at 0.04 mg/l for less than 2 hr per day. Copper concentrations at this station were as high as 0.08 mg/l and may have affected the results with the snails.

Basch and Truchan (62) and Truchan (63) have described field studies conducted at five power-generating stations in Michigan. Caged brown trout were held for 96 hr in the intake and discharge channels. Most chlorinations were for 30 min. Intermittent concentrations of TRC of 0.14 to 0.17 and 0.18 to 0.19 mg/l for two and four 30-min chlorinations, respectively, were lethal to 50 percent of the trout in 48 hr. In 96 hr, 50 percent of the trout were killed at TRC concentrations of 0.02 to 0.05 and 0.17 and 0.18 mg/l for three and six 30-min chlorinations, respectively. Caged brown bullheads, fathead minnows, and various species of sunfish were not killed at these same concentrations. Underwater observations of resident fish in distress were made at two plants at concentrations ranging from 0.2 to 0.5 mg/l.

Marcy (64) in his study of vulnerability and survival of Connecticut River fish entrained at a power plant, observed no deaths due to chlorination. He concluded that, since only one-fourth of the system was chlorinated at a time, the resultant dilution and additional chlorine demand lowered the TRC concentration below an effect level. The residual chlorine was recorded at less than 0.1 mg/l.

Biological studies at the Quad-Cities Nuclear Power Station (65) resulted in the conclusion that the periodic concentrations of chlorine in the heated effluent appeared to have a more significant impact on the biotic communities in the Mississippi than the increased water temperature. Productivity of periphytic algae was reduced at times downstream from the plant.

Fox and Moyer (66) determined in situ productivity in the discharge to the Gulf of Mexico by the fossil-fueled Crystal River plant. Primary production was decreased an average of 57 percent by plant passage and chlorination. In the absence of chlorine the average decrease was 13 percent. Each of eight intake pipes was chlorinated to a TRC concentration of 0.1 to 1.0 mg/l with dilution by unchlorinated water from the other seven units. Consequently, TRC was never detected (limit was 0.01 mg/l) at the discharge where productivity was reduced.

Polgar et al. (67) described a model for entrainment loss of zooplankton in a power-generating station in the lower Potomac River. Experimental data on zooplankton (46) were compared with the results of this model. This comparison showed that radical depletions in the population of copepodite and naupliar stages occurred that could not be accounted for by cooling system or delayed entrainment deaths.

The effect on yellow perch, coho salmon, rainbow trout, alewife, and spottail shiner of a single 30-min exposure to various concentrations of TRC

was evaluated by Seegert et al. (68). After exposure the fish were placed in unchlorinated water and observed for as long as 48 hr. The LC50 values for yellow perch tested at 10, 15, and 20° C were 7.7, 4.0, and 1.1 mg/l, respectively. All other species were tested only at 10° C, and they were all more sensitive than the yellow perch. The LC50 value for coho salmon was 1.25 mg/l; for rainbow trout, 2.0 mg/l; for alewife, 2.25 mg/l; and for spottail shiner, 3.2 mg/l. Later studies (69) resulted in LC50 values for alewife at 15, 20, and 25° C of 2.3, 1.65, and 0.92 mg/l, respectively. For coho salmon, the LC50 values at 15 and 20° C were 1.35 and 0.92 mg/l, respectively. Additional tests with yellow perch at 25° C provided an LC50 of 1.0 mg/l and with rainbow trout at 15° C the LC50 value was 0.9 mg/l. Delayed mortalities occurred in most of the exposures and in the tests with the alewife 45-95 percent of the TRC was free chlorine. Yellow perch and rainbow trout were also exposed for a single 5-min period. Only one yellow perch out of 100 was killed at TRC concentrations between 9 and 27 mg/l; only two out of 130 rainbow trout were killed at concentrations from 1.5 to 4.0 mg/l. Brooks and Seegert (70) exposed rainbow trout and yellow perch to three 5-min doses of TRC separated by 3-hr recovery periods and observed their survival for 48 to 72 hr. At 10° C the LC50 value for the rainbow trout was 2.87 mg/l and for the yellow perch at 10° C the LC50 value was 22.6 mg/l as TRC. Mortalities were delayed for both species usually occurring 2 to 24 hr after the third exposure.

Bass and Heath (71, 72), Bass et al. (73), and Bass (74) exposed rainbow trout to intermittent chlorinations three times per day. Breathing, coughing frequency, and heart beat were measured. During each chlorination blood pO_2, pH, and heart rate decreased, whereas breathing and coughing rates increased. Increased mucus production and damage to respiratory epithelium were apparent in histological sections of gill tissue. The authors concluded that the primary mode of action of chlorine is through gill damage leading to death by asphyxiation. Bass and Heath (75) and Heath (76) also exposed bluegills to intermittent chlorinations simulating the operation of steam electric generating plants. Small bluegills were exposed to four concentrations of TRC (0.21 to 0.52 mg/l) at four temperatures (6 to 32° C). No deaths occurred at 0.21 mg/l at any temperature, and only a few deaths occurred at 0.31 mg/l. A concentration of 0.52 mg/l caused 50 percent mortality in less than 75 hr; high temperatures resulted in more rapid death.

Warren, in addition to the continuous exposures discussed earlier (38), also has begun studies on intermittent chlorination with predominantly free chlorine and cutthroat trout. In preliminary experiments single 15- to 60-min exposures resulted in few deaths at TRC concentrations up to 1.0 mg/l.

Dickson and Cairns (77) conducted 74 static bioassays with goldfish at different exposure times (15 min to 24 hr), exposure frequencies (one to six times), chlorine species (free and combined chlorine), sizes of test fish, and temperatures (a range of 12.5 to 16.5° C and a range of 17 to 22.5° C). Temperature and size of fish had no detectable effect on toxicity. When the concentration of TRC and the duration of an exposure were held constant, a doubling of the frequency of exposure from one to two increased the predicted fish mortality by a factor of 8. Combined chlorine appeared in these tests to be slightly more toxic than free chlorine. The TRC continuous-exposure 24-hr LC50 for the goldfish was 0.27 mg/l. The authors concluded that a chlorination protocol of two to three exposures of

15 to 30 min per day with a TRC concentration of 0.5 to 0.75 mg/l would not be acutely lethal to fish equally as sensitive as or less sensitive than the goldfish. Unfortunately, as indicated elsewhere in this review, the goldfish is one of the fish species more resistant to TRC.

Collins (78) exposed emerald shiners to single, 30-min doses of TRC and observed their behavior and survival for 96 hr. At 10 and 25° C the LC50 values for juveniles were approximately 1.4 and 0.3 mg/l, respectively. He also observed that the juveniles were less sensitive than the adults at 10° C.

Stober and Hanson (79) exposed juvenile chinook salmon to five TRC concentrations in sea water at four temperatures and four exposure times between 7.5 and 60 min. Juvenile pink salmon were tested at three temperatures. Dynamics of chlorine decay in sea water were also discussed. A decrease in the tolerance of both species was demonstrated with increasing temperature and exposure time. Mean times to equilibrium loss and death were presented. The result of studies with fluctuating TRC concentrations during a 2-hr chlorination under continuous-flow conditions with juvenile pink salmon was an LC50 value of 0.045 mg/l; under these conditions, the time for 50 percent of the test fish to die occurred in about 100 min at approximately 0.5 mg/l.

The interaction of temperature shock and up to 10-min exposures to TRC concentrations in sea water of 0.3 to 0.5 mg/l was studied by Hoss et al. (80). Larval flounder exposed to 0.3 mg/l with no temperature shock survived for up to 5 min, but after 7 min the exposure mortality was 20 percent, and after 10 min mortality was 100 percent. At 0.3 mg/l there was no loss of larval menhaden after 7 min when there was no temperature shock. At 0.5 mg/l the mortality of these fish was 100 percent after 10 min. Between 40 and 60 percent of juvenile mullet died when exposed to 0.3 mg/l for 7 to 10 min with no thermal shock. When these three fish species were subjected to shock temperature increases of as much as 12° C, their chlorine sensitivity increased greatly, indicating at least an additive effect of the two stresses.

Margrey et al. (81) exposed Atlantic menhaden and hogchoker to concentrations of chlorine between 0.125 and 2.0 mg/l under continuous and intermittent exposure conditions for 96 hr or until 100 percent mortality occurred. The Atlantic menhaden was the more sensitive species and, in general, the fish survived longer when exposed intermittently rather than continuously.

Early developmental stages of plaice and Dover sole were exposed under continuous-flow conditions to TRC in sea water by Alderson (82). The eggs of plaice were most resistant with 72- to 192-hr LC50 values of 0.64 and 0.105 mg/l, respectively. The early larval stages of both fish species were most sensitive with 48- to 96-hr LC50 values between 0.025 and 0.071 mg/l. Metamorphosed fish were of intermediate sensitivity with 96-hr LC50 values for TRC between 0.070 and 0.095 mg/l. No attempt was made to differentiate between the effects of the various chlorine and bromine compounds produced as a result of sea-water chlorination. The author concluded that in sea water the TRC probably consisted mainly of bromine compounds.

Nash (83) has presented some of the problems and uses of condenser cooling water for fish farms. Specific problems resulting from chlorination of these waters are discussed in relation to water chemistry.

The effects of intermittent chlorination on marine and freshwater invertebrates are less well understood than the effects on fish. McLean (84) exposed colonies of hydroids attached to suspended nylon lines to TRC concentrations of 1.0 to 4.5 mg/l for 1 and 3 hr and then returned the colonies to their natural habitat. Differences in exposure time caused no significant difference in new growth. The percentage of new growth was slightly lower in colonies subjected to concentrations of 2.5 to 3.5 mg/l than in the controls. McLean (85) also exposed barnacle larvae, copepods, grass shrimp, and two species of amphipods to TRC in the laboratory in continuous-flow test chambers. Most of the exposures were for 5 min to a TRC concentration of 2.5 mg/l, after which the test organisms were transferred to chlorine-free water for observation. Average mortality was 80 percent for barnacle larvae and 90 percent for copepods exposed for 5 min. Mortality was low in the shrimp and amphipods exposed to TRC for 5 min. When the exposure time for these two species was increased to 3 hr, mortality of one amphipod species immediately after the 3-hr exposure was about 25 percent and after 96 hr mortality was 97.2 percent. Shrimp deaths were comparable after the 3-hr exposure, and almost all were dead at 96 hr.

Acute bioassays were conducted (86) to determine the effect of thermal shock and chlorine exposure on the estuarine copepod, Acartia tonsa. Laboratory conditions simulated power plant operation with an 8° C thermal shock and brief chlorinations. A single dose of chlorine was added to static test chambers, and after 96 hr a TRC concentration of 0.75 mg/l measured at the start of the test was fatal to 30 percent of the copepods. At 1.15 mg/l all copepods died. Latimer (87) and Latimer et al. (88) exposed two species of freshwater copepods to 30-min chlorinations at several temperatures. Based on the added chlorine and the ammonia content of the test water, the authors expected that the resulting TRC would be predominantly free chlorine. As temperature increased over the range of 10 to 20° C, sensitivity also increased, and the 30-min LC50 values for Cyclops bicuspidatus thomasi decreased from 14.68 to 5.76 mg/l. The 30-min LC50 values for Limnocalanus macrurus at 5 and 10° C were both 1.54 mg/l. The two species had entirely different regression slopes. Cyclops was able to withstand a much wider range of TRC concentrations than was Limnocalanus. Twenty-five percent of Cyclops died at concentrations below 5 mg/l, whereas 25 percent were able to survive a concentration of 45 mg/l.

Protozoan communities were exposed to free chlorine three times in a 2-hr period by Cairns and Plafkin (89). The number of species (relative to controls) decreased significantly at concentrations above 1.15 mg/l. Free chlorine concentrations above 0.66 mg/l, when added every 20 min over the 2-hr period, caused a significant decrease in the number of species surviving. Certain species appeared to be more tolerant of the chlorine stress, and these species were among those previously known to be tolerant of other stress conditions. In these experiments an increase in the number of chlorine additions over the 2-hr period effectively lowered the concentration that produced a significant reduction in species diversity. Changes in species numbers or species diversity were not different from those expected as purely additive effects.

Ginn et al. (90) studied the survival of estuarine amphipods entrained in condenser cooling water from the Hudson River. Mean survival at the intake and discharge stations with no chlorination ranged from 90 to 99 percent; during chlorination mean survival at the two discharge stations ranged from 51 to 81 percent. Data on TRC concentrations were not presented.

Davies and Jensen (91) examined quantitatively the entrainment of zooplankton at representative thermal power plants operating in a once-through mode on a river, lake, and estuary. Thermal and chlorination effects were both evaluated. The TRC concentrations between 0.25 and 0.75 mg/l at the Indian River plant (estuarine) did not reduce the zooplankton motility ratio by more than 50 percent. In two of three instances when chlorine levels of 1.00 mg/l were measured at the Indian River plant and when concentrations from 0.50 to 5.0 mg/l were recorded at the Chesterfield Plant (river), the zooplankton motility ratio was reduced by 85 to 100 percent. The plant sited on a freshwater lake used mechanical cleaning instead of chlorination.

Stage I larvae of the American lobster (less than 48 hr old) were exposed by Capuzzo et al. (92) to 30- and 60-min exposures at three temperatures to free and combined chlorine in sea water filtered through activated charcoal to remove dissolved organics and a 1-μ filter to remove particulates. The larvae were observed during a 48-hr period. Applied chloramine was more toxic than corresponding concentrations of applied free chlorine with estimated *LC50* values at 25° C of 16.3 mg/l (applied concentration of free chlorine) and 2.02 mg/l (applied concentration of chloramine). Toxicity was reduced at 20° C and increased at 30° C. Approximately 18 percent of the applied concentration of free chlorine and chloramine was detectable, amperometrically, as residual chlorine. No reason for this low recovery in filtered sea water was determined. Respiration rates of the lobster larvae were reduced even 48 hr after the short exposure periods at all tested concentrations. Similar results were obtained at 0.05 mg/l applied chloramine and 0.10 mg/l applied free chlorine.

One significant problem in antifouling control is the significant lack of experimental data with which to determine the actual need for chlorination in terms of frequency and duration of use and the times of years when antifouling is necessary. It is readily apparent that in many cases chlorine is used in great excess. For example, during 1973 to 1975 Commonwealth Edison Company (with plants on and around Lake Michigan) has achieved a system-wide 30 percent net reduction in chlorine use (93). This reduction represented a savings of 1.5 million pounds of chlorine in this one system.

Carpenter and Macalady (94) discussed the ability of various analytical methods for the determination of residual chlorine in sea water. They concluded that present methods do not appear to be satisfactory if close compliance with regulatory statutes involves analysis of samples from wastewater treatment plants and electric generating stations. They attribute this problem to the lack of understanding of exactly what occurs when sea water is chlorinated, and this knowledge of the chemical species is a prerequisite to designing proper analytical methods.

Eppley et al. (95) studied the decline in free and residual chlorine in filtered, non-filtered, and ultraviolet-oxidized sea water. In the filtered and non-filtered samples there was a rapid initial decline followed by a slower decline; no decline was observed in the ultraviolet-oxidized sample probably because of removal of those molecular species that react with chlorine. In addition, they observed decreases in marine phytoplankton photosynthesis after intermittent exposure to power plant chlorination as low as 0.01 mg/l of TRC. The authors discussed the analytical problems unique to sea water as influenced

by bromine and the formation of residual bromide after the initial formation of hypobromous acid and hyprobromite. They suggest that differences between toxicity between freshwater and marine situations might reflect differences in the toxicity of residual chlorine vs. residual bromine.

DECHLORINATION

Many of the above recent toxicity studies have also evaluated the use of dechlorination for the elimination of TRC toxicity by using a variety of chemicals and procedures such as lagooning of wastewater (19, 20), sodium bisulfite (26, 27), sulfur dioxide (28, 29), sodium sulfite (46, 51), activated carbon (52), and sodium thiosulfate (24, 62, 63). Tsai (10) included a brief summary of several studies that involved dechlorination of wastewaters. Carbon filtration alone may not always reduce TRC to acceptable levels for toxicity testing (45).

Martens and Servizi (96) studied the influence of dechlorination of primary sewage with sulfur dioxide on toxicity, pH, and dissolved oxygen. The pH and dissolved oxygen were the same whether or not dechlorination was used. In addition to the elimination of chlorine toxicity, the toxicity of the chlorinated-dechlorinated primary effluent was less than the toxicity of untreated primary effluent, suggesting degradation of some toxic constituents by the chlorination-dechlorination treatment of primary effluent. No acute effects of a mean sulfur dioxide residual of 2.20 mg/l were detectable after 96 hr. When the study plant (Lulu Island Treatment Plant) reaches the design flow of 16 mgd, the cost of dechlorination is estimated to be about one-third the cost of disinfection with chlorine.

Photochemical action for the removal of free chlorine from water before its use in fish tanks has been discussed by Armstrong and Scott (97). Commercial mercury-lamp water sterilizers can be used. The treated water appeared to be harmless to fish.

Shifrer *et al.* (98) evaluated the toxicity of the effluent from a small physical-chemical waste treatment system at a Utah highway rest station. Sodium sulfite was used to eliminate TRC toxicity so that less toxic components of the effluent could be evaluated.

AVOIDANCE

This interesting phenomenon has been studied to only a slight degree recently. Meldrim *et al.* (99) determined behavioral avoidance responses to temperature and chlorine under various conditions of salinity, light, and dissolved oxygen concentration over the annual range of temperature in the Delaware River estuary. Responses to various TRC concentrations were determined for the white perch, mummichog, hogchoker, grass shrimp, sand shrimp, and blue crab. Measurements of free and combined chlorine indicated that most of the TRC was free chlorine. Results of multiple regression analysis indicated that only

the bluecrab preferred potentially lethal concentrations. In addition, avoidance concentrations were generally inversely related to temperature and to light level.

Bogardus et al. (100) described a new avoidance test chamber and determined the avoidance responses of the mimic shiner, river shiner, and the bullhead minnow. The system was buffered to pH 8.6 with an excess of ammonium chloride to ensure that monochloramine was the principal component of TRC in this study. Test concentrations were analyzed amperometrically and ranged from 0.005 to 0.5 mg/l. Each group of fish was exposed to a low concentration, and if no response was detected, the concentration was raised to the next level. The procedure was continued until avoidance was observed. The mimic shiner was the most sensitive and was able to detect and avoid all tested concentrations between 0.005 and 0.45 mg/l. River shiners avoided test solutions when the monochloramine concentration reached 0.15 mg/l. The avoidance-inducing concentration for the bullhead minnow was between 0.03 and 0.05 mg/l. Even after the test ended and no TRC could be detected, the mimic shiner still avoided entering the effluent side of the test chamber. Observations for up to 30 min indicated that this minnow had learned to avoid the toxicant side of the chamber. At higher concentrations the avoidance reaction was so great that even when the observer placed his hands in the chamber, the fish could not be driven into the toxicant side.

The study by Tsai (20) previously described was not designed to detect or observe avoidance, but the study did relate the presence and absence of 45 fish species in relation to measured TRC concentrations. Unless there were additive or synergistic effects of TRC and such factors as ammonia concentration, turbidity, detergents, or other major components of domestic wastewaters, it may be that Tsai (20) was actually observing avoidance. Tsai and Fava (101) and Fava and Tsai (102, 103) studied the avoidance of chlorinated sewage effluent, chloramine solutions, and free chlorine solutions by the blacknose dace, a small minnow. The fish avoided a TRC concentration of 0.51 mg/l. after 20-min exposures to the chloramine solutions and to the chlorinated sewage. The avoidance concentration for free chlorine was 0.92 mg/l. The threshold avoidance concentrations, indicating an initial slight response to TRC, were 0.13, 0.18, and 0.61 mg/l for chlorinated sewage, chloramine, and free chlorine, respectively. When the exposure time was increased to 220 min the fish avoided all the chloramine and free chlorine solutions at concentrations as low as 0.07 mg/l. Unchlorinated sewage effluents were not avoided. The authors also observed that some fish definitely preferred concentrations of free chlorine at 0.52 and 0.19 mg/l after a 20-min exposure. The authors concluded that chloramines are the major constituents in chlorinated sewage effluents causing fish to avoid these effluents and that this avoidance response explains the absence of fish in streams below outfalls when there is no evidence of fish kills.

FORMATION OF CHLORINATED ORGANIC COMPOUNDS

Among the factors contributing to the present concern for the potential environmental impact of wastewater chlorination is the formation of chlorine-containing organic constituents in chlorinated effluents. Jolley (104, 105) described a method to study this potential problem by combining radioactive tracer chlorination in the laboratory with high-resolution chromatography. Using this procedure he detected over 40 chlorine-containing compounds in primary domestic effluent. Subsequent studies (106) dealt with secondary effluent. These

studies also attempted to estimate the concentrations of the chlorine-containing compounds. The qualitative results with secondary effluent were comparable to those with primary effluent (105). The actual concentrations of these compounds ranged from 0.00024 to 0.0043 mg/l. Jolley et al. (107) have described their analytical procedures in detail.

Glaze et al. (108) studied secondary effluent before and after laboratory chlorination for 1 hr and have presented some preliminary results. Mass spectroscopy and mass spectroscopy-gas chromatography were used after neutral organic constituents were concentrated by adsorption on resin. The gas chromatography studies with a Coulson detector demonstrated concentrations of chlorine compounds in the range of 0.001 to 0.05 mg/l in the wastewater effluent. Only a few of the compounds the authors observed were identified, but chloroform was positively identified. In addition, Glaze and Henderson (109) and Glaze et al. (110) collected grab samples of secondary municipal effluent, and superchlorinated them (>1,000 mg/l) with a contact time of 1 hr. Organics were resin-extracted and analyzed. Qualitative and quantitative results were obtained for a few dozen chemicals with most concentrations in the range of 0.010 to 0.040 mg/l. The highest concentration detected was 0.285 mg/l for 3-chloro-2-methylbut-1-ene.

Jolley et al. (111) have also studied the formation of chloro-organics by chlorination of cooling waters. Cooling water was chlorinated in the laboratory for 75 min at 2.0 mg/l. Over 50 chlorine-containing organic compounds were found. They estimated that several hundred tons of chlorinated organics are produced annually in the United States by this antifouling process. The concentrations of chloro-organic products formed during chlorination of cooling waters and process effluents were estimated by Jolley et al. (112). Concentrations ranged from 0.0002 to 0.02 mg/l for phenols, purines, aromatic acids, pyrimidine, and nucleoside.

The ability of natural humic substances to act as precursors for haloform production during chlorination of natural waters was studied by Rook (113). Four haloforms were clearly recognized; these were chloroform, dichlorobromomethane, chlorodibromomethane, and bromoform. Further experimentation with a non-polluted natural water taken from a lake in a peaty region provided the same result; chloroform production attained a concentration of 0.2 mg/l. An unexplained phenomenon was the formation of bromohaloforms in proportions much greater than the ratio of added chlorine to bromine concentrations in the river water.

A report by Carlson et al. (114) dealt with the aqueous chlorination of aromatic systems and the examination of the relationship of chlorine incorporation to pH and contact time. The extent of chlorine incorporation into the biphenyl nucleus (chlorobiphenyl production) was pH dependent, and above pH 4 extended reaction times were required to observe extensive chlorine incorporation. The ability of the relatively unactivated biphenyl nucleus to be chlorinated under a wide range of aqueous chlorination conditions, plus the facile chlorination of those compounds (phenols and aromatic ethers) that resemble a variety of naturally occurring compounds, suggests that many chlorinated organics could be produced in this manner.

Carlson and Caple (115, 116) found that chlorine reacted readily in an aqueous medium with a variety of compounds (α-terpineol, oleic acid, abietic acid, cholesterol, etc.) that are known to be present in wastewaters subjected to

chlorine-renovation processes. Biological effects on Daphnia magna, a cladoceran, of the compounds formed by chlorine incorporation were also examined. Toxicity generally increased with increasing chlorine content. The effects of chlorination on biological oxygen demand (BOD) were examined by comparing the BOD requirements of a sample containing a given chemical system with that of its chlorinated products. The results indicated that the chlorinated material is generally degraded to a lesser degree than the parent compound and that the lowered BOD values appear, at least in the case of phenols, to be due to the increased toxicity of the chlorinated material to the degrading species.

The study of chloro-organic formation has also extended into the area of biological studies. Kopperman et al. (117) have studied the residues accumulated by fish exposed to chlorinated secondary wastewater for their entire life cycle through reproduction. The exposure tests with these fish were described earlier (29). By using gel permeation chromatography for tissue cleanup and gas chromatography-mass spectroscopy techniques for identification, various compounds were identified that were either not present in fish reared in non-disinfected effluent or were present in much lesser quantities. Some of these compounds were di- and tri-chlorophenol, di- and tri-chlorobenzenes, and trichloroanisole. Tribromoanisole was tentatively identified in fish raised in chlorobrominated effluent. The principal concern about these chloro-organics as they relate to fish residue is that the incorporation of chlorine into an organic molecule increases its lipophilic character, which usually results in increased toxicity and bioaccumulation.

Fourteen industrial organic chemicals were studied during biological wastewater treatment in semi-continuous activated sludge systems (118). The ability of these chemicals to combine with free chlorine was then determined. Five of the initial compounds combined readily with chlorine under conditions commonly used in effluent chlorination. The ability of these compounds (phenol, m-cresol, hydroquinone, aniline, and dimethylamine) to react with chlorine can be related to the structural characteristics of the chemicals. Several of the reaction products were examined in respirometer studies and found to be resistant to degradation by a heterogenous microbial population. In addition, static 96-hr bioassay tests with the fathead minnow were conducted to determine the toxicity of five reaction products. The range in LC50 values was from 0.01 to 10 mg/l. Continuous-flow aquaria with a plant-animal ecosystem were also tested with trichlorophenol, benzoquinone, and trichloroaniline.

The formation of chlorinted compounds during wastewater chlorination is not unique. Unbleached pulp typically becomes fully bleached in a process that involves one or more chlorinations with hypochlorite and chlorine dioxide (119). The toxicity of five compounds, separated in a pure state from the effluent, was determined with rainbow trout and static bioassay procedures. The TRC after the chlorination stage was 50 to 130 mg/l. The compounds and their 96-hr LC50 values were: 3,4,5-trichloroguaiacol, 0.75 mg/l; 3,4,5,6-tetrachloroguaiacol, 0.32 mg/l; monochlorodehydroabietic acid, 0.6 mg/l; dichlorodehydroabietic acid, 0.6 mg/l; and 9, 10-epoxystearic acid, 1.5 mg/l. These same compounds were found in effluents collected from six other kraft mills.

Gehrs et al. (120) studied the toxicity to carp eggs of two reaction products of wastwater chlorination: 4-chlororesorcinol and 5-chlorouracil. Eggs that were water-hardened before exposure were more resistant to these

chemicals than were non-water-hardened eggs. Both reaction products reduced hatchability at nominal concentrations as low as 0.001 mg/l. Subsequent studies (121) with 5-chlorouracil have been conducted to study its sublethal effect on carp. Nine replicated concentrations (0.001 to 10 mg/l) were tested; solutions were changed twice daily during the 3-day incubation period before hatching. The effect end point was the percentage of larvae that were malformed just after hatching occurred. The percentage of malformed carp larvae increased with increasing concentrations of 5-chlorouracil above 0.5 mg/l.

Kopperman et al. (122) determined the toxicity to Daphnia magna of 14 phenolic compounds having the ability to incorporate chlorine over a wide range of pH and concentration. Toxicity increased significantly with increased halogen content.

Katz and Cohen (123, 124) tested several classes of common compounds before and after excessive chlorination (5 to 10 mg/l) to determine whether such treatment would affect the toxicity of those compounds. Mosquitofish was the test fish for the static bioassays. The toxicity of phenol and tryptophan was increased by chlorination, whereas the toxicity of tannic acid was decreased. Chlorination had no influence on most of the tested compounds, which included urea, ornithine, histidine, uracil, linear alkene sulfonate, leucine, and others.

Morris (125) concluded that the reactions of aqueous chlorine in water chlorination are not indiscriminate and unpredictable. In fact, they follow quite well-defined pathways in accord with general principles of organic reaction mechanisms. He added that it is generally possible to predict something of the nature and extent of the reactions of aqueous chlorine with components of water and wastewater.

The Conference on the Environmental Impact of Water Chlorination was held at the Oak Ridge National Laboratory, Oak Ridge, Tennessee, October 22-24, 1975. The major objective of its program was to present and discuss the best available data concerning the formation and effects of chlorinated organic compounds associated with the use of chlorine as a biocide or in process treatment. Three major technical sessions dealt with the aqueous chemistry of chlorine, biomedical effects of chloro-organics, and environmental transport and effects. Several presented papers are discussed in this review and the proceedings of this conference will be published.

The chlorination of municipal water supplies, although not directly appropriate to this review since such water has no contact with aquatic life, does result in the formation of chlorinated organic compounds. Such information is useful to those concerned with this problem in sanitary wastewaters and cooling waters and will be discussed briefly. Morris and McKay (126) have critically reviewed the available literature on formation of halogenated organic compounds during the chlorination of water supplies. They concluded that chlorination is not indiscriminate and does not lead to the formation of a wide variety of chlorinated derivatives with any and all organic pollutants. Chlorination results in a limited number of well-defined reactions on a few specific types of organic structures. The identifiable initial reactions are electrophilic aromatic substitution of positive chlorine and electrophilic addition of positive chlorine to appropriately activated double bonds.

Volatile organics were collected from New Orleans drinking water and pooled human blood plasma by Dowty et al. (127). Using gas chromatography and mass spectrometry, they identified 13 halogenated hydrocarbons in the drinking water and five in the blood plasma. Tetrachloroethylene and chloroform were found in both plasma and water. The relative concentrations of the halogenated hydrocarbons in the drinking water varied considerably from day to day. The gas chromatographic procedures used for this study have been described (128). Additional studies by Dowty et al. (129) have identified approximately 70 organic constituents in finished water, the preponderence of which are either aromatic or halogenated aliphatic and aromatic. The authors noted that essentially every compound observed in finished water was present, usually in a much lower quantity, in the precursor river water. They assumed that trace quantities of such compounds may have originated from wastewater treatment plant discharges upstream.

Stevens et al. (130) studied the halogenation of organic compounds during chlorination of drinking water. The major known products of these reactions are trihalomethanes. Several factors influence production of these compounds; they are precursor compound concentration, pH, type of disinfectant used (free or combined chlorine), and temperature. In addition, the point of chlorination in the treatment process is also a significant factor in trihalomethane production. The latter is the most important variable that could be used to reduce the concentrations of these chlorinated compounds in drinking water.

MISCELLANEOUS

In reality, chlorine is a pesticide and it is used principally to control a variety of human pests (bacteria, slimes, algae, setting organisms, and viruses). The old adage "If a little will do it, a lot will do it better" applies to chlorine use just as it does to pesticide use. Although the usual desired TRC concentration at the discharge is 0.5 to 2.0 mg/l, concentrations frequently exceed those necessary for microbiological control. Snoeyink and Markus (4) measured TRC below 20 wastewater treatment plants in central Illinois and determined that monochloramine was the predominant species and that free chlorine concentrations generally were very small. The TRC concentrations were as high as 5.17 mg/l in grab samples. The authors' measured concentrations were in general much higher than the values reported by the treatment plant personnel, who used the orthotolidine method. Most of the plants used chlorination equipment that added chlorine at continuous rates regardless of chlorine demand or wastewater flow through the plant. This procedure would obviously result in overchlorination at any times other than the maximum daily loading. McKersie (5) found concentrations as high as 10.3 mg/l in effluent from 22 wastewater treatment plants in southern Wisconsin. Simultaneous testing with an amperometric titrator and the chlorine comparators used by the plant operators showed consistent inaccuracy with the colorimetric comparators. As in the Illinois plants, the chlorinators at most of the Wisconsin plants studied were set at one rate for each 24-hr period. As expected, the TRC concentrations increased with decreasing wastewater flow.

It is very likely that if chlorinators were operated automatically by chlorine demand or a monitored TRC concentration in the discharge, the reduction in the amount of chlorine used would be significant economically as well as environmentally. This reduction would eliminate much of the TRC discharged to surface waters.

Pereira et al. (131) developed a predictive model for free residual chlorine concentrations. The model considers hydrodynamic transport, chlorine-demanding reactions, and gaseous exchange with the atmosphere in a flowing stream. Draley (132) measured free chlorine and TRC in the circulating-water system of a power plant cooling tower. For a typical chlorination period the concentration of free chlorine at the condenser was about 0.1 mg/l whereas the TRC concentration reached a maximum of 0.63 mg/l at the condenser discharge. More than 2 hr elapsed, after the addition of chlorine was stopped, before TRC was no longer detectable (about 0.01 to 0.02 mg/l). The data for the buildup of TRC were fitted with a kinetic expression. Good fits, with reasonable values for the constants, indicated that the model was correct. A model for gradual decay of combined chlorine also appeared reasonable in that, again, good fits for the data occurred. Lietzke (133) reviewed the literature to obtain available kinetic and thermodynamic data on the various chemical reactions of chlorine in order to develop a model for predicting the composition of chlorinated water discharged from power plant cooling systems. Future refinements of this model will incorporate temperature-dependent expressions for the various rate and equilibrium constants.

Nelson (134) also developed and analyzed a mathematical model to predict TRC in cooling tower blowdown at any time during the chlorination cycle. The model and its eight variations were useful in predicting TRC concentrations in the blowdown. The model also permitted alterations in existing chlorination schedules, which minimized chlorine waste and reduced chlorine concentrations in the blowdown. As a result of concentration of dissolved materials in power plant blowdown, the amperometric titration method had interferences (135). The addition of sodium pyrophosphate as a complexing agent removed those interferences caused by iron and copper in the water matrix.

Marinenko (136) and Marinenko et al. (137) developed a new monitor for the National Bureau of Standards to detect low concentrations of TRC. Iodine, which results from oxidation of potassium iodide by TRC, was measured amperometrically in a system in which coulometrically generated iodine is used as a system calibrant. Laboratory and field tests with this monitor and other instruments were performed for intercomparison. The portable monitor was capable of measuring TRC concentrations as low as 0.001 to 0.003 mg/l.

Johnson (138) expressed concern about the inability to selectively measure the various species of chlorine in order to minimize the concentration of chlorine necessary to produce a microbiologically acceptable water. He compared present field, laboratory, and continuous methods for free and combined chlorine residual for specificity, reagent stability, accuracy, and simplicity. These methods were the acid orthotolidine methods, DPD, SNORT, LCV, FACTS, amperometric titration, copper-gold amperometric cells, and the NBS flux monitor. He also evaluated a new analytical method specific for HOCl or NH_2Cl in the presence of the poor disinfectants OCl^-, organic chloramines, and other interferences.

A water pollution investigation (139) of the Fields Brook, Ashtabula River and Harbor (Ashtabula, Ohio) revealed high concentrations of chlorine, using the orthotolidine method, resulting from industrial discharges. Measurements of total residual chlorine in Fields Brook varied from approximately 1 to 12 mg/l near its mouth. One measurement of 35 mg/l was recorded at an outfall. Concentrations were reduced by dilution in the Ashtabula River to 0.03 to 0.05 mg/l.

Aquatic populations were greatly reduced but may have been the result of high concentrations of other toxic materials.

Chlorination of urban water supplies has been found responsible for two epidemics of acute hemolytic anemia in uremic patients undergoing hemodialysis (140). Chloramines produced denaturation of hemoglobin by their direct oxidizing capacity and their ability to inhibit red cell reductive metabolism. In relation to this problem of methemoglobin production in human blood by chloramine, Grothe and Eaton (141) exposed two groups of fathead minnows to a potentially lethal concentration of monochloramine (1.5 mg/l) and compared the methemoglobin concentrations in their blood with those in control fish. After a 1-hr exposure the concentrations of methemoglobin in the blood of monochloramine-exposed fish were 32.0 and 29.4 percent of total hemoglobin, and in the control fish the concentrations were 2.8 and 2.6 percent of total hemoglobin. The authors concluded that lethal concentrations of chloramine apparently kill fish by virtue of their powerful ability to oxidize hemoglobin and cause death by anoxia when the red cells can no longer deliver an adequate oxygen supply to the tissues.

Studies (45) described earlier with aquatic invertebrates used a chlorine-dosing system designed to control intermittent and periodic additions of chlorine. This dosing system has been described in detail by Gregg and Heath (142). It is basically a timer-operated pump set to simulate the chlorination schedule of many steam-electric generating plants.

Cairns et al. (143, 144) suggested that, since chlorine causes the epithelium of fish gills to slough off, and a copious amount of mucus is produced which clogs the gill lamellae, asphyxia may be a major cause of death resulting from exposure to TRC. Consequently, they hypothesized that elevated temperatures, which would lower dissolved oxygen and increase the respiratory demand for oxygen, probably would increase the toxicity of chlorine to fish.

Katz (145) studied the influence of sodium, potassium, calcium, and magnesium on the acute toxicity of chlorine to mosquitofish under static bioassay conditions using tap water dechlorinated by direct sunlight, deionized water, and several concentrations of sea water. Free and combined residual chlorine were measured during the exposures by the orthotolidine and sodium arsenite methods. Sodium at concentrations between 500 and 3,000 mg/l most effectively reduced the toxicity of 0.5 and 1.0 mg/l chlorine and prolonged survival time of the test fish. Lower sodium concentrations did not influence toxicity, and mortality at higher concentrations of sodium probably reflected osmotic stress. When the mosquitofish were acclimated to 25 percent sea water before transfer to tap water with chlorine added, they were significantly more resistant to the chlorine than fish acclimated to tap water. When 100 mg/l of calcium was added to tap water or when equivalent amounts of both calcium and sodium were added to distilled water, chlorine toxicity was similar to an exposure to chlorine in tap water, which indicates that calcium nullified the protective action of the sodium ion. This ionic interplay with chlorine was highly suggestive of some membranal effect. Katz (145) also described several studies that evaluated the influence of chlorine on gill membrane permeability. Sodium efflux and gain in fish weight were a direct consequence of the addition of chlorine. The author suggested that ammonia excreted by fish gills rapidly converts free chlorine to chloramines so that the fish would be exposed mainly to combined chlorine. A model was presented,

based on the results of these studies, for the action of chlorine in the gill of freshwater fish. Consequences of physiological effects at the membrane and environmental levels were discussed.

Yearling coho salmon were exposed for 12 weeks to chlorinated sewage plant effluent diluted with seawater under continuous flow conditions (146). Concentrations of effluent of 1.1 and 3.6 percent (0.009 and 0.03 mg/liter of TRC, respectively, resulted in reductions of hemoglobin and hematocrit to levels indicative of anemia. Observations of the erythrocytes revealed lysed and degenerating cells, increased numbers of circulating immature cells, and abnormal cells. The highest tested no-effect concentration of TRC was 0.003 mg/liter (0.3 percent effluent).

AQUATIC LIFE CRITERIA AND APPLICATION FACTORS

Various problems arise during the development of aquatic life criteria. One principal difficulty is evaluating the quality of the available data so that their influence on the eventual recommendations is proportional to their importance and accuracy. As discussed earlier, the analytical method used in various studies can have a great influence on the accuracy of the result , whether chemical or biological. However, unless the investigator specifically compares two or more methods, one of which is amperometric, there is no way to measure the accuracy of his results. No quality control procedure can be applied to data generated years ago. Consequently anyone attempting to summarize and evaluate research data before developing a recommendation or criterion should accept the results of most analytical methods for TRC but the possible limitations of the data should not be forgotten.

Criteria for freshwater aquatic life have been recommended (9) for both continuous and intermittent exposures of TRC. Criteria were recommended for both coldwater fish species and warmwater fish species, a distinction that is becoming less justifiable because of the sensitivity of various minnows (18, 25, 52, 75). For continuous and intermittent exposures to TRC the criteria for trout and salmon and most aquatic organisms were 0.002 and 0.04 mg/l, respectively. For warmwater fish species and less sensitive aquatic species the criteria were 0.01 (continuous) and 0.2 mg/l (intermittent). The criteria for intermittent exposures to TRC were applicable for up to 2 hr/day. Basch and Truchan (147) developed comparable TRC criteria for freshwater fish. Their criteria for coldwater fish species were 0.005 mg/l for continuous exposure and 0.04 mg/l for intermittent exposure. For warmwater species the criteria were 0.02 and 0.2 mg/l for continuous and intermittent exposure, respectively. The National Academy of Sciences (148) in its compilation of freshwater aquatic life criteria recommended a single maximum TRC concentration of 0.003 mg/l at any time or place. They also concluded that aquatic organisms can tolerate short-term exposure to higher concentrations. Consequently, it was also recommended that when intermittent chlorination is used TRC should not exceed 0.05 mg/l for no longer than 30 min in any 24-hr period. For marine aquatic life the National Academy of Sciences (148) recommended an application factor of 0.1 and 96-hr LC50 data from sea-water bioassays for the most sensitive species to be protected. They also suggested that free residual chlorine in sea water in excess of 0.01 mg/l can be hazardous to marine life.

As discussed earlier, Mattice and Zittel (58) plotted available data on median effect concentrations of TRC on freshwater and marine organisms and estimated acute and chronic toxicity thresholds for each group. These thresholds represent a dose-time combination below which there are either no deaths (acute threshold) or no effect, no matter how long the exposure (chronic threshold). The freshwater and marine acute thresholds are different, but both are time dependent. The chronic toxicity thresholds are constant and for freshwater and marine organisms are 0.0015 and 0.02 mg/l, respectively. As a result of apparent basic differences in sensitivity of freshwater and marine organisms, which may be the result of different TRC chemistry, the time at which the chronic thresholds begin are different. For freshwater organisms the chronic threshold begins at nearly 1,000 min of exposure, whereas for marine organisms it begins at just over 100 min. The authors indicated that these thresholds were for protection of aquatic life; the implication of a no-effect chronic threshold for continuous exposure to chlorine seems consistent with the purpose of developing aquatic life criteria. The freshwater threshold (0.0015 mg/l) is similar to the criteria for all freshwater aquatic life discussed earlier (0.002 mg/l (9), 0.003 mg/l (148), and 0.005 mg/l (147)).

The European Inland Fisheries Advisory Commission (149) recommended criteria for European freshwater fish species. They attempted to convert most published data on TRC toxicity to free chlorine, and the recommended criterion was 0.004 mg/l of free chlorine. Total residual chlorine criteria were temperature and pH dependent, but not ammonia dependent, and ranged from 0.004 to 0.121 mg/l. They concluded that chlorine is too reactive to be very persistent in most streams, and the upper limit for fish survival could be set closer to lethal levels since avoidance is likely to provide additional protection at higher concentrations. They tentatively suggested that the criterion be 0.004 mg/l of free chlorine, which should result in few or no fish deaths.

A concept commonly used for the development of criteria for freshwater fish is the application factor. An application factor is the ratio between an experimentally derived safe concentration of a toxicant and the 96-hr LC50. Application factor criteria for freshwater aquatic life have been recommended (148) for such materials as copper, nickel, zinc, un-ionized ammonia, free cyanide, linear alkylate sulfonates, and phenolics.

Safe and 96-hr LC50 concentrations of residual chlorine for freshwater fish were reported in three different studies. Arthur and Eaton (150) in studying the effect of chloramines on the fathead minnow observed a safe concentration for all life stages, including reproduction, of 0.0165 mg/l. The 96-hr LC50 calculated with their data was 0.120 mg/l. The application factor would then be 0.14. Arthur et al. (28) used chlorinated secondary wastewater effluent and also determined safe and 96-hr LC50 concentrations of TRC for the fathead minnow; the concentrations were 0.014 and 0.108 mg/l (means 0.130 and 0.086 mg/l), respectively. The application factor would be 0.13. Ward et al. (29) also used chlorinated secondary effluent and the fathead minnow. Their safe and 96-hr LC50 concentrations were 0.010 and 0.089 mg/l, respectively. The individual LC50 values were 0.095 and 0.082 mg/l. Again, with their data the application factor would be 0.11. The mean of these three application factors is 0.13, or approximately one-eighth.

Of the three chronic studies with fathead minnows (28, 29, 150) two (28, 29) concluded that growth or survival was as sensitive as or more sensitive than reproduction in estimating the chronic safe concentration. If we assume, therefore,

that growth is a sensitive indicator of chronic TRC effects on fish, the data of Warren (38) may be used to calculate an application factor also. He has conducted several growth studies with coho salmon exposed to chloramines. Growth was inhibited at TRC concentrations between 0.020 and 0.024 mg/l; no effects on growth were detected at 0.010 to 0.012 mg/l. The mean of his 96-hr LC50 values was 0.077 mg/l. When we use 0.011 mg/l as an estimate of the safe concentration for coho salmon, the application factor, based on coho salmon data, is 0.14. The mean application factor derived from the three fathead minnow chronic tests is 0.13.

The purpose of the application factor (151) is to estimate safe concentrations for fish species that cannot, or probably would not, be tested chronically in the laboratory. A ninety-six-hr LC50 value for a specific toxicant is determined for such a species, and this value is multiplied by the application factor for that toxicant in order to estimate safe concentrations for chronic exposure of the species to the toxicant in question. If we use the 96-hr LC50 values for TRC for the most sensitive freshwater fish species such as trout, salmon, and minnows and multiply these values by one-eighth, we are estimating their approximate safe concentrations for chronic exposure to TRC. Most of the 96-hr LC50 values for these more sensitive fish species range between 0.040 and 0.080 mg/l. The estimated safe concentrations of TRC would therefore range between 0.005 and 0.010 mg/l. The minimum estimate of 0.005 mg/l is consistent with those criteria discussed earlier that range from 0.002 to 0.005 mg/l. It is obvious that an application factor of one-eighth (0.13) will provide no "margin of safety" in that it results in a criterion that is very close to a lethal concentration. Mattice and Zittel (58) concluded that a barely non-lethal concentration of TRC can be estimated by multiplying the 96-hr LC50 by 0.37, or about one-third. Since one-third of a 96-hr LC50 is an estimate of an acute, non-lethal concentration of TRC and one-eighth of a 96-hr LC50 is an estimate of a chronic, safe concentration of TRC, the narrowness of the lethal-to-safe range is of significant concern.

The U.S. Environmental Protection Agency, in its compilation of water quality criteria (152), has proposed aquatic life criteria for chlorine. For salmonid fish the criterion was 0.002 mg/l, and for marine and other freshwater organisms the criterion was 0.010 mg/l. These criteria, if approved, will be sent to the States, who will consider them in the process of developing proposed State standards for receiving waters. The data summarized in this review would support a single criterion for TRC of 0.003 mg/l for freshwater aquatic life continuously exposed to TRC. The more recent data on warmwater fish species necessitates this change from the two-criteria concept.

Criteria for intermittent exposure of aquatic organisms to TRC should be time related and those developed by Mattice and Zittell (58) are appropriate at this time.

REGULATIONS

The U.S. Environmental Protection Agency recently proposed (153) an amendment of the Secondary Treatment Information regulation contained in 40 CFR Part 133 and promulgated pursuant to Section 304(d)(1) of the Federal Water Pollution Control Act Amendments of 1972. Section 301(b)(1)(B) of this act required that effluent limitations, based on secondary treatment, be achieved on all publicly owned treatment works in existence on July 1, 1977, or approved for a construction grant prior to June 30, 1974. The proposed amendment deleted the fecal coliform bacteria limitations from the definition of secondary

treatment. An agency task force (154) reviewed the U.S. Environmental Protection Agency policy on wastewater disinfection and the use of chlorine. It became evident that present disinfection policy inadvertently encouraged the use of chlorine to control bacteria since nearly all facilities were using chlorine or chlorine-based compounds. In some instances the present policy required disinfection even when it was unnecessary. Now the U.S. Environmental Protection Agency intends that disinfection only be considered when public health hazards need to be controlled. The exclusive use of chlorine for disinfection should not be continued where protection of aquatic life is of primary consideration. Alternate means of disinfection and disinfectant control (dechlorination) must be considered where public health hazards and potential adverse impact on the aquatic and human environments co-exist. Disinfection should not be required where no benefits accrue.

SECTION VI

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16. ABSTRACT

The literature since 1972 pertaining to wastewater and cooling water chlorination is discussed under the following headings: review papers, chlorinated municipal wastewaters, continuously chlorinated water, intermittently chlorinated water, dechlorination, avoidance, formation of chlorinated organic compounds, aquatic life criteria and application factors, and regulations.

Field and laboratory research results support a single criterion of 0.003 mg/l for continuous exposure of freshwater organisms. The former distinction between warmwater and coldwater systems is no longer appropriate; recent data indicate that several freshwater fish species are as sensitive as trout and salmon.

The present concern for the formation of chlorinated organics in water and wastewaters is justifiable and the greatest present need is to determine the ecological significance, if any, of these results. The future course of wastewater chlorination will be greatly influenced by the recent proposed changes in the Environmental Protection Agency's regulations on secondary treatment. The changes intend that disinfection only be considered when public health hazards need to be controlled, and that the exclusive use of chlorine should not be considered where protection of aquatic life is of primary consideration. Where these uses co-exist, alternate means of disinfection must be considered.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS		b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Aquatic animals	Condenser tubes	DESCRIPTORS (continued)	6C, 6F, 6I, 6T, 7B, 7C
Aquatic plants	Cooling towers		
Bioassay	Cooling water	Fouling prevention	
Biocides	Dechlorination	Ozone	
Bromination	Disinfectants	Ozonization	
Bromine halides	Disinfection	Sewage	
Chlorination	Fishes	Sewage treatment	
Chlorine organic compounds		Toxicology	

18. DISTRIBUTION STATEMENT	19. SECURITY CLASS *(This Report)*	21. NO. OF PAGES
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