Transactions of American Fisheries Society, Vol. 57-58, 19

Pb in Fish - Aquatic Life 1524

TOXICITY EXPERIMENTS WITH FISH IN REFERENCE TO TRADE WASTE POLLUTION

DAVID L. BELDING

From the Evans Memorial and Boston University School of Medicine

I. THE PROBLEM OF WATER POLLUTION

Of the various problems confronting our fisheries, the one which demands the most immediate attention is the water pollution. If allowed to increase, it means the serious depletion and even ruin of a great part of our inland and coastal fisheries, and the longer it remains unregulated, the more difficult will become its ultimate control. Already it has made such inroads that the restoration of many heavily polluted streams is practically impossible. The water pollution evil is not a mere local issue; its scope is national, and its elimination requires concerted action. The battle for unpolluted streams must be fought simultaneously throughout the country by a well organized campaign.

OBSTACLES.—The elimination of water pollution means overcoming many obstacles, especially in organizing a co-operative campaign among states where local conditions, different laws, and divergent forms of pollution present numerous complications. The antagonism of the manufacturing interests must be overcome and indifferent public opinion aroused by a campaign of education. Laws must be carefully drawn and must be sufficiently broad in their application, as well as rigid in their enforcement to facilitate the handling not only of transitory pollution, but also of cases where polluting material is surreptitiously emptied into a stream under the pretense of a nominally operating disposal system. The present lack of definite knowledge as to the quantity and kind of pollution which will injure fish life is a serious handicap. Since the cost of freeing badly polluted streams in heavy manufacturing centers is prohibitive, it may be advisable to allow them to continue to serve as sewers for waste disposal, but in border line cases which demand a careful consideration of all conditions, proper standards for determining whether or not a stream should be cleared of pollution are badly needed.

EFFECT ON FISH LIFE.—Two kinds of water pollution affect fish life, sewage and trade wastes. Sewage enters the stream from private toilets and cesspools, and in the form of effluents from municipal systems. Owing to its prominence as a public health factor, methods of disposal have been devised by which

27-1928, pp. 100-119

it will eventually be controlled. ~~If unregulated, it~~ damages water supplies and manufacturing interests, endangers public health through contamination of edible shellfish, and is injurious to fish environment. Trade wastes include all forms of waste material from industrial sources. Fish preservation is concerned chiefly with this important type of pollution, which directly and indirectly destroys fish life.

Acting directly, pollution reduces the oxygen supply, renders fish unfit for food, drives them from their habitat, and causes death or predisposition to disease. Indirectly, it may cause destruction of eggs and young, reduction of the spawning grounds, changes in bottom and vegetation, and limitation of food supply. The effect of severe recent or transitory pollution in flagrant cases is conspicuously indicated by the presence of dead or dying fish, but unappreciable continuous pollution may be fully as harmful due to a cumulative effect over a long period.

Remedial Measures.—Constructive work, in order to avoid strong opposition, must be undertaken gradually. In the first place, new pollution must be prevented; secondly, small streams with few sources of pollution must be cleared; and finally, through the cooperation of manufacturers, the pollution in the larger streams must be eliminated or diminished. The manufacturers can be influenced by the diplomatic but forceful handling of each case. As a last resort in refractory cases where moral suasion is of no avail, rigid enforcement of laws must be invoked. To insure such action simple, direct, readily enforceable laws are necessary. Favorable public opinion is absolutely necessary for the successful enforcement of laws. It may be obtained by making an appeal through the medium of pamphlets, posters, magazine and newspaper articles, and lectures on the basis of fair play, loss of outdoor enjoyment, and danger to public health.

Investigations.—The ultimate solution of the problem of water pollution involves certain biological investigations: (1) Suitable means must be found for eliminating various types of trade wastes. (2) Standards for judging as to when a stream is polluted must be determined. These standards must be applicable to different types of streams and to various kinds and concentrations of water pollution, in order to ascertain the maximum amount each stream may handle with safety. (3) The direct and indirect effect of the various constituents of water pollution upon fish life must be observed.

Since the factors involved in these biological problems are so complicated, the immediate and ultimate effect of various types of trade waste pollution must be carefully studied before we shall be in a position to determine the exact amount of pollution fish can survive or to formulate standards for judging the amount of pollution any given stream can handle.

II. FACTORS INFLUENCING EXPERIMENTAL STUDIES UPON THE EFFECT OF WATER POLLUTION ON FISH

The chief obstacle in obtaining accurate data relative to the effect of various inorganic and organic wastes upon fish life is the difficulty of controlling the various complicating factors which enter into an experiment. Unless their influence is recognized, errors of interpretation are inevitable. As a rule the conditions, under which these experiments are made, are far from ideal. Field tests must be performed in an environment in which exact laboratory technique is impossible. Necessarily, all experimental work of this nature should be most throughly scrutinized before acceptance.

1. FISH

The selection of the fish for experimental work is of primary importance. All possible safeguards should be taken to secure healthy, normal individuals, since disease and weakness render them susceptible to changes in environment.

INDIVIDUAL VARIATION.—Individual fish, particularily if diseased or weak, vary considerably as to the lethal dose, and unless an ample number of suitable controls are used results may prove untrustworthy. Even healthy fish of the same size, weight, and appearance may show several hours differance in the onset of symptoms and time of death.

FAMILY.—Certain families are more resistant than others, e. g., Cyprinidae vs. Salmonidae. Hardy fish exist where the more delicate cannot survive. With unfavorable changes in environment, the more susceptible fish are the first to perish, although if destruction through natural agencies is limited to a habitat frequented by certain species, it may appear otherwise. Certain families may be resistant to one form of trade waste pollution and susceptible to another, e.g., perch and bass. Obviously, it is impossible to make adequate comparisons as to the toxic effect of various chemicals unless tests are made with the fish from the same family. Even then the lethal dose for one species may not correspond to that for another.

SPECIES.—A difference in susceptibility between members of the same family is shown by the action of copper sulphate upon Chinook salmon, brook trout, and rainbow trout, which were held in the same pool. The solution which was sufficient to cause the death of all the salmon and seriously effect the brook trout, producing marked symptoms and causing a few deaths, had no apparent effect upon the rainbow trout. The well known resistant qualities of the rainbow trout and the susceptibility of the salmon hold true in protection against chemical poisons.

AGE.—Observations upon fingerlings and adult brook trout indicate that there is a difference between young and old in their resistance to disease and to toxic substances. Brook trout fry are relatively immune to infection with *B. salmonicida*, whereas adult fish are extremely susceptible. Fry in the yolk sac stage withstand certain trade wastes better than older fry or adult fish. Young fish are more resistant than older fish to some chemicals and less resistant to others, e. g., fingerling trout appear more resistant to carbolic acid, a corrosive poison, and more susceptible to hydrochloric acid than adult trout.

SIZE AND WEIGHT.—The difference in the resistance of old and young fish may be due in part to age and in part to size, weight, and greater surface area which produce greater demands upon the body metabolism. A comparison of the oxygen requirements of forty-one 19 cm. and eighteen 26 cm. trout with the same combined weight illustrates the latter point. The larger trout showed earlier symptoms of distress and consumed during the first two hours nearly twice the quantity of dissolved oxygen used by the smaller fish.

VITALITY.—Experimental results depend upon the general condition of the fish. Vitality varies at different seasons. It is lowest immediately after spawning, during extremely warm weather, and during periods of food scarcity. Although fish reared under artificial conditions have not the vitality of the wild stock, they undergo confinement better and are preferable for experimental work of this character. The active, darting, nervous species tend to perish when closely confined.

II. ENVIRONMENT

WATER.—The constituents of the water may alter the effect of certain chemicals, e. g., the lethal dose of copper sulphate has been observed by Marsh to vary in different waters from 1:400,000 to 1:6,500,000. In certain waters the presence of carbonic acid may change ammonia to the less toxic ammonium carbonate, sulfids may be more quickly oxidized to sulphates, and buffer action may influence the hydrogen-ion concentration. The waters favorable to fish life will produce a more sturdy species than an unfavorable environment.

Toxicity experiments in a new environment are apt to record an abnormal susceptibility on the part of the test fish, e. g., the transfer of alewives or even the hardy bullhead to cold spring water rapidly lowers their resistance.

CONTAINERS.—The ratio of the size and number of the fish to the container is important. Goldfish, which are accustomed to aquarium life, may be successfully tested in small containers for long periods, whereas large fish die rapidly because of oxygen depletion, increased carbonic acid, and injury in the cramped quarters.

FLOW OF WATER.—Tests with chemicals are most satisfactory when made without change of water. The solution should be of uniform strength before the fish are added. If tests are made with flowing water some method of producing a constant uniform concentration must be devised. Mechanical injury, such as forcing weakened fish against the screen at the lower end of the pool must be avoided.

OXYGEN.—The amount of dissolved oxygen in the water has a direct bearing on toxicity tests. Each species of fish requires a minimum oxygen level for existence and a higher one for normal existence. Unless the surface area of the container is sufficient to supply the necessary oxygen during the course of the experiment, the influence of diminished oxygen must be counteracted by some form of aeration. With trade wastes which have an oxygen demand sufficient to reduce the dissolved oxygen below the point of tolerance, it is difficult to separate the effect of oxygen depletion from chemical poisoning, e. g., with hydrogen sulfid, the dissolved oxygen is decreased with the concentration of the gas and a double effect is produced, hydrogen sulfid poisoning and diminished oxygen.

TEMPERATURE.—Cold water fish can stand a comparatively high temperature when the change is gradual, but sudden changes may prove destructive. Brook trout are killed by a rapid transfer from 55 to 80 degrees Fahrenheit. Extreme fluctuation in night and day temperature has been observed to cause a high death rate in brook trout fingerlings. Increased temperature, by lowering the vitality of the fish and favoring the growth of the infecting organism, plays an important role in fish epidemics. Similarly it produces more rapid destruction of fish by chemical pollution; e. g., the lethal dose of hydrogen sulfid for carp at 40 degrees Fahrenheit was 34 p.p.m. and at 65 degrees 6 p.p.m.

III. TOXICITY STATISTICS

These variable factors indicate the difficulty of obtaining accurate results in field and laboratory studies of the effect of chemical wastes upon fish and explain the discrepancies in the results of different investigators. They emphasize the importance of carefully controlled tests and the necessity of determining the minimum lethal dose of each chemical for various species of fish in order to establish standards.

Our laboratory and field experiments deal with a single phase of the pollution problem, the immediate effect of certain chemical constituents of trade waste pollution upon young and adult fish. Observations are limited to the symptoms and mortality occurring within twenty-four hours. Although these crude tests determine the concentration which fish may withstand for twenty-four hours, they give no information as to the effect of weaker solutions acting over a longer period of time. For the sake of completeness we have compared our results with the statistics of previous investigators.

METHODS OF WORK.—The chemicals used in these tests comprised acids, bases, salts, and gases, either constituents of trade wastes or remedies used empirically by fish culturists in the treatment of fish diseases. Unless otherwise specified, the chemicals were of high grade purity. Brook trout, rainbow trout, chinook salmon, carp, goldfish, and suckers were used. Owing to their sensitivity to changes in environment brook trout proved the most satisfactory indicators, although carp and goldfish were more convenient to handle. For most tests two to three year old trout averaging about 26 cm. in length and 215 grams in weight were used. Carp, suckers, and goldfish were employed for tests with hydrogen sulfid and paper mill wastes.

The trout were held without change of water in four wooden tanks containing 780, 646, 147, and 147 liters respectively. Five adults were placed in the larger tanks and two in the smaller. Control tests showed that this number survived without discomfort in these tanks for longer than twenty-four hours. The temperature of the water ranged from 53 to 60 degrees Fahrenheit, averaging 56. In a few instances tanks which received a slow seepage of spring water were used. Gasoline, petrol, leather board, paper mill, and wool scouring waste tests were conducted in tanks and in Chase hatching jars. For laboratory experiments with sodium and hydrogen sulfid, 50 liter glass aquaria supplied with air forced through a Berkefield filter were used. In field work with paper mill waste and hydrogen sulfid, large wooden wash tubs proved convenient.

SYMPTOMS.—Chemical poisons produce certain general symptoms, but the various substances differ qualitatively as to selectivity of symptoms and quantitatively in the rapidity of onset and course of development. The general type and specific characteristics of the chemical determine the symptomatology. At times the selective action of the poison is sufficiently defined as to be of diagnostic value. The early symptoms are more important from the standpoint of differential diagnosis, since the late symptoms are usually confused with those present in all dying fish.

In order to be available for ready reference, the characteristic symptoms produced by twice the minimum lethal dose have been tabulated for the various chemicals (Table 1). A description of the general symptoms in chronological order is given first, and under the individual chemicals, attention is called to those of diagnostic importance.

1. IRRITATION.—The first noticeable sign after the fish is immersed in the chemical bath is increased activity in the form of swimming, wavy movements and rubbing the body against sides of the tank. The degree of activity and rapidity of onset varies with the chemical. With the more irritating chemicals the fish becomes covered with a grayish mucus, evidently a protective reaction. Its absence in the salmon may explain the relative susceptibility of that species.

2. INACTIVITY.—The period of activity is followed ordinarily by a period of sluggishness, in which the fish lies inactive at the bottom of the tank. Response to stimulus during

this stage is particularly poor and even during the active stage is below normal.

3. SWIMMING.—The fish first shows restlessness by incessant movement through the water. At times it apparently tries to get out of the noxious environment. Then its movements become more irregular and it aimlessly wanders around the tank, frequently striking the sides. It may glide quickly to the surface and occasionally leap from the water. Intervals of resting at the bottom and surface swimming follow, the movements becoming more and more incoordinate.

4. OXYGEN HUNGER.—Oxygen hunger is manifested by surface swimming and gulping air. It is due either to lack of dissolved oxygen or to interference with gill function.

5. EQUILIBRIUM.—With the approach of death the fish loses its powers of equilibrium. Suddenly it turns on its side and swims with a convulsive flutter. Sometimes it is able to right itself, but more often there is increasingly feeble and spasmodic swimming on the back with slow irregular respirations.

6. RESPIRATIONS.—The respiratory movements of normal trout range from 90 to 100, averaging about 95. Owing to individual variation, it is difficult to determine accurately changes in respiration. With some chemicals there is a temporary increase in the respiratory rate, with others no change or a slight decrease; but with the onset of marked symptoms of discomfort and with the approach of death practically all fish manifest some form of slow convulsive respiratory movement.

LETHAL DOSE.—The minimum lethal dose for the various chemicals is given in Table 2. It represents the lowest dilution capable of producing death within twenty-four hours. For comparison the statistics of other investigators have been included.

1. HYDROCHLORIC ACID.—With minor differences hydrochloric, nitric, and sulphuric acids produce the same general type of symptoms in trout and differ chiefly in respect to the lethal dose. The effect of an acid upon fish depends both upon ionization and upon inherent toxic properties. The toxic dilution of these acids ranges between a pH of 4 and 5, yet fish have been found in waters as acid as pH 4.6.

Hydrochloric acid is used in plating and in the manufacture of chemicals and dyes. The characteristic symptoms are gradual onset, early surface swimming, lack of response to stimuli, and a mucous coating. There is terminal loss of equilibrium, irregular slow respiration, and frenzied rushing. The minimum lethal dose is 1:100,000, which is stronger than that obtained by other investigators.

2. NITRIC ACID.—Nitric acid is used in the manufacture of fertilizer, chemicals, munitions, etc. It acts more rapidly than hydrochloric and suggests the action of toxic factors other than ionization. The chief characteristics are marked irritation and activity, rapid formation of mucus, decreased forceful convulsive respiration, early loss of equilibrium, and frenzied rushings, particularly in the terminal stages.

3. SULPHURIC ACID.—Sulphuric acid is used in wool scouring, nail and iron works, and in the manufacture of munitions, chemicals, and starch. The onset of symptoms is slower, activity is less noticeable, and sluggishness is more marked than with hydrochloric and nitric acid. There is no frenzied rushing or convulsive actions. The fish, covered with mucus, swim slowly and blindly around the pool aimlessly bumping the sides.

4. ACETIC ACID.—The organic acids present greater diversity of action than the inorganic acids. Acetic acid is used by fish culturists in the treatment of diseased fish. Trout can survive for a short time immersion in a dilution of 1:200. The symptomatology presents no characteristic features; slow onset, moderately increased activity followed by sluggishness, and the formation of a protective covering of mucus.

5. CARBOLIC ACID.—Carbolic acid is characterized by its corrosive action. The most striking feature is its rapid and severe irritating effect even in high dilutions. The fish immediately endeavor to rub off the irritating substance by rolling over on the bottom and by rubbing against the sides of the pool. They show no evidence of oxygen hunger and seek the corners of the pool. In dilute solutions the fish soon lose their equilibrium and lie on their back or side for several hours before death with spasmodic twitching of head and tail. The respiratory movements are slightly increased, become irregular, forceful, and jerky. Dying trout show a general congestion of gills, liver, and viscera.

6. Tannic Acid.—Tannic acid, an astringent derived from galls and closely related to gallic acid, is used in the manufacture of ink and leather. Trout gradually become uneasy, swimming restlessly to the surface with oxygen hunger and then lie quietly on the bottom with respiration noticeably quickened. Increasing signs of discomfort terminate in erratic swimming, surface jumping, loss of equilibrium, and death. The rapid respiratory rate and oxygen hunger indicate injury to the gills.

7. Ammonium Hydroxide.—The action of the three alkalis, ammonium, potassium and sodium hydroxide, differs only in degree of toxicity. Ammonium hydroxide is a product of gas and ammoniacal works, the effluents of which have been found to kill fish. The symptoms are rapid and fulminating. The fish dash wildly about, jumping and thrashing at the surface. With the minimum toxic dose signs of irritation appear in two and one-half hours and in three hours the fish turn on their backs with altered respiration and spasmodic struggling. Respirations are markedly decreased and death occurs after five hours. On removal to fresh water dying fish recover in six hours. The symptoms increase with the strength of the solution. In ten times the toxic dose there is immediately frenzied rushing, jumping out of the tank, and swimming on back in 15 minutes. The hydroxide is changed to the less injurious carbonate by free carbonic acid, and therefore the action of this chemical varies in different waters.

8. Sodium Hydroxide.—Trade wastes containing sodium hydroxide are derived from a number of industrial processes. It produces an irritating effect upon the fish which rapidly become covered with mucus. In strong solutions the movements of the fish become sluggish. It comes to the surface gasping and blindly bumps the sides of the tank. In the minimum lethal solution it remains at first quietly at the bottom and then comes to the surface gasping. Later it lies on its back with periods of rapid dashing and leaping. The strong dilutions rapidly inhibit the respiration, but with the minimum lethal dilution no change is noticed for the first two hours, then the respiratory movements become slow and feeble and after the loss of equilibrium show marked irregularity.

9. Potassium Hydroxide.—Potassium hydroxide produces the same train of symptoms as sodium hydroxide, namely, sluggishness, irritation and distress, oxygen hunger, decreased respiration and loss of equilibrium. With solutions of similar

strength the action is less marked than that of sodium hydroxide.

10. ARSENATE OF LEAD.—The particular preparation used contained 12.5 per cent of As_2O_5, 31.5 per cent of Pb O_2, 0.75 per cent of water soluble arsenic and moisture not over 50 per cent. It is used for spraying trees. Its slow action produces no characteristic symptoms in fish.

11. CALCIUM HYPOCHLORITE.—The toxic action of calcium hypochlorite depends upon the free chlorine and therefore is transitory. With strong solutions the fish pass at once into a stage of irritation, followed by sluggishness and lack of response to stimuli. With dilute solution the onset is insidious and the two stages occur together. The respiratory movements are increased. The fish rubs its body against the sides of the pool and swims to the surface with increasing frequency, which finally culminates in rushing and leaping at the surface and spasmodic gasping. With loss of equilibrium the fish rests head down in the water, turning with a top-like motion. Then it settles to the bottom where it repeats from time to time the rushing tactics which each time become more feeble. The smaller trout were less affected than the larger. The extreme oxygen hunger, surface rushing, and peculiar head balancing are of diagnostic importance.

12. COPPER SULPHATE.—Copper sulphate is used as an algacide for water supplies. Owing to the extreme range of toxicity for different species of fish and for various waters, it is difficult to determine the quantity which may be used for algacidal purposes without injury to fish life. The fish first shows increased activity and then lies sluggishly on the bottom with increased spasmodic respiration. Occasionally it darts through the water rapidly, usually inclined slightly to the side. Loss of equilibrium follows, the fish remains quiet and unresponsive to stimuli, and occasionally struggles to the surface. The effect of the poison persists even after transfer to fresh water which does not save the life of the fish.

13. FERROUS SULPHATE.—Ferrous sulphate occurs in wastes from iron and nail works. With the exception of Clark and Adams, observers find that iron salts have a relatively low toxicity. There is nothing characteristic about the symptoms, which develop slowly.

14. MERCURIC CHLORID.—This chemical is used as an antiseptic. It acts as a tissue coagulant. At death the aur-

icles are filled with blood and the gills pale. The fish, apparently unaffected at first, die suddenly without noticeable symptoms.

15. POTASSIUM PERMANGANATE.—Fish culturists use this antiseptic for the treatment of diseased fish. Fish immersed in strong solutions recover promptly when placed in fresh water. The skin acquires a yellow tinge from the chemical and the gills are deeply injected. The chief symptoms are surface swimming and leaping.

16. HYDROGEN SULFID.—This gas produces in man acute and fatal poisoning in chemical works and sewers through its action on the respiratory tract. It is one of the most toxic of the coal tar wastes. It is associated with decomposition of wood pulp and is formed in small amounts in nature. Since potassium and sodium sulfid hydrolize into hydrogen sulfid in the blood their action is similar. Small amounts are without perceptible effect but larger quantities paralyze respiration. Since it is oxidized rapidly in the body it has no cumulative action. The limit of tolerance of different species of fish has a wide range. Its action depends upon the concentration of the gas and the temperature of the water.

The chief symptoms are irritation, frenzied rushing, respiratory paralysis, partial recovery of respiration, stupor, loss of equilibrium, oxygen hunger, and recovery on removal. The respiratory changes are diagnostic. There is an initial lowering or suspension of respiratory movements followed by a partial recovery and subsequently a decline in rate with increasing irregularity.

TRADE WASTES.—The lethal dose of the trade wastes varies with their composition and dilution. In addition to their chemical constituents certain wastes tend to reduce the supply of dissolved oxygen in the water.

The sulphite liquor from paper pulp manufacture has a slow action. It produces no special symptoms beyond those usually observed in dying fish. Leather board waste acts in a similar manner. With wool scouring waste, which usually contains sulphuric acid and organic debris the fish manifests uneasiness, endeavors to leap out of the tank and shows oxygen hunger.

The writer wishes to acknowledge the able assistance of Mr. J. A. Kitson in conducting the field experiments.

TABLE 1

SYMPTOMS

	INORGANIC ACIDS			ORGANIC ACIDS			ALKALIS				SALTS				GASES	
	Hydrochloric Acid	Nitric Acid	Sulphuric Acid	Acetic Acid	Tannic Acid	Carbolic Acid	Ammonium Hydroxide	Potassium Hydroxide	Sodium Hydroxide	Arsenate of Lead	Calcium Hypochlorite	Copper Sulphate	Ferrous Sulphate	Mercuric Chloride	Potassium Permanganate	Hydrogen Sulfid
1. Onset																
Rapid		+				+	+				+	+				+
Slow	+		+	+	+			+	+	+			+	+	+	
2. Irritation																
Restlessness	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Rubbing movements		+				+					+					
Secretion of mucus	+	+	+	+				+	+							
3. In activity																
Sluggish movements	+	+		+	+	+		+	+	+	+	+	+	+		+
Lack of response to stimuli	+		+	+		+					+	+				+
4. Swimming																
Rapid movements	+	+					+				+	+			+	+
Irregular movements	+	+	+	+	+	+	+	+	+		+	+				+
Frenzied rushing and leaping		+			+	+	+				+				+	+
5. Equilibrium																
Early loss		+				+	+				+					+
Late loss	+		+	+	+			+	+	+		+	+	+	+	
6. Respiration																
Increased					+	+					+	+			+	
Decreased	+	+	+				+	+	+	+						+
Normal				+									+	+		
7. Oxygen Hunger					+		+	+	+		+					+

TABLE 2

LETHAL DOSE OF CERTAIN CHEMICALS FOR FISH

Chemical	Authority	Species	Dilution: Killed within 24 hours	Dilution: No effect
I Inorganic Acids Hydrochloric	Belding	Brook trout	160,000	
	Clark & Adams	Carp, shiners, suckers	274,000	
	Marsh	Mummichogs, sunfish	125,000*	
	Penny & Adams	Minnows, goldfish		50,000
Nitric	Belding	Brook trout	640,000	
	Penny & Adams	Minnows, goldfish	50,000*	
	Clark & Adams	Carp, goldfish, suckers,		50,000
		shiners		174,000
Sulphuric	Belding	Brook trout	160,000	
	Penny & Adams	Minnows	50,000*	
		Goldfish		50,000
	Wells	Blue-gill sunfish	40,800	277,000
	Marsh	Mummichogs and sunfish	83,000*	
II Organic Acids Acetic	Belding	Brook trout	20,000	
	Penny & Adams	Minnows	8,750	
		Goldfish	3,500*	
Tannic	Belding	Brook trout	160,000	
	Penny & Adams	Minnows	14,000*	
		Goldfish	7,000	
Carbolic	Belding	Brook trout	160,000	
	Weigelt	Trout	200,000*	
	Penny & Adams	Minnows	70,000*	
		Goldfish	3,000*	7,000
III Alkalis Ammonium Hydroxide	Belding	Brook trout	160,000	
	Clark & Adams	Carp, shiners, suckers	77,000*	106,000
	Weigelt	Small fish	33,000*	
Potassium Hydroxide	Belding	Brook trout	20,000	
	Penny & Adams	Goldfish	7,000*	35,000
		Minnows	35,000*	
	Wells	Blue-gill sunfish	18,000	36,000
Sodium Hydroxide	Belding	Brook trout	40,000	
	Clark & Adams	Carp, shiners, suckers	14,000	18,000
IV Salts Arsenate of Lead	Belding	Brook trout	40,000	
Calcium Hypochlorite	Belding	Brook trout	Over 100,000	
	Mason	Trout	2,000,000	
	Waring	Tench	200,000*	
		Trout	125,000*	
Copper Sulphate	Belding	Brook trout	250,000	
	Moore & Kellermann	Trout		7,000,000
		Black bass		500,000
	Penny & Adams	Minnows, goldfish	100,000*	200,000
Ferrous Sulphate	Belding	Brook trout	7,500	
	Clark & Adams	Carp, shiners, suckers	157,000	200,000
	Mason	Trout	10,000	20,000
	Penny & Adams	Minnows & goldfish	10,000	

*Time not specified.

DUP050312906

TABLE 2. (Continued.)

LETHAL DOSE OF CERTAIN CHEMICALS FOR FISH

	CHEMICAL	AUTHORITY	SPECIES	DILUTION: KILLED WITHIN 24 HOURS	DILUTION: NO EFFECT
	Mecuric Chloride	Belding	Brook trout	80,000	
		Mason	Trout	20,000	
	Potassium Permanganate	Belding	Brook trout	160,000	
V	Gases Hydrogen Sulfid	Belding	Carp	160,000	
			Suckers	263,000	
			Brook trout	1,163,000	
			Goldfish, aquarium	233,000	
			Goldfish, wild	40,000	
		Shelford	Carp	300,000	
VI	Trade Wastes		Sunfish	187,000	
			Trout	100,000	
		Weigelt	Tench	10,000	
	Leather Board	Belding	Brook trout	500	
	Paper Mill Sulphite liquor	Belding	Brook trout	500	
		Marsh	Perch & bass		200
		Knight	Trout, perch, rock bass	10	
		Thomas	Trout	750*	3,000
	Wool Scouring	Belding	Brook trout	1,000	

*Time not specified

BIBLIOGRAPHY

Clark, H. W. and Adams, G. O.
Ann. Rep. Mass. State Board of Health, 1912, p. 366.

Knight, A. P.
Supplement 32nd Ann. Report, Dept. Marine and Fisheries, Canada, 1902.

Marsh, M. C.
U. S. Geol. Survey, Water Supply Papers 181-192, 1907.

Moore G. T. and Kellermann, K. F.
U. S. Dept. Agriculture, Plant Ind. Bull. 1903, 76.

Penny and Adams
See Rafter, G. W. and Baker, M. N., Sewage Disposal in the U. S., p. 344.

Thomas, A.
Trans. Am. Fish. Soc., March 1919.

Waring, G. E. Jr.
Modern Methods of Sewage Disposal.

Weigelt, C.
See "Water Supply"—Mason, Wm. P.

Wells, M. M.
Biol. Bull. 1915, 29, p. 221.

Discussion

MR. J. B. DOZE (Kansas): Did I understand you to say that the action of copper sulphate on the perch and on the trout was similar, that the trout would stand more than the scaled fish?

DR. BELDING: I have only used it on trout and salmon. The figures which I gave on perch, bass, sunfish, etc., were furnished by Moore and Kellerman back in 1905; and I believe Marsh also has certain figures showing the variation of different species. In the case of perch the dilution was one and one-half millions, and Marsh claims that in the case of trout it should be seven millions.

MR. DOZE: Carp?

DR. BELDING: In the case of carp he gave three millions; but I will be perfectly frank and say that I question its correctness.

MR. DOZE: I was going to question that, because we sometimes kill carp with it when it does not affect the bass. I questioned that particular experiment.

DR. BELDING: It is not my work; I simply quoted it.

DR. FIELD: In experiments made with the State Board of Health of Massachusetts on Lake Massapoag for a determination of the quantities of copper sulphate, when one part to three million was used, I found that 550,000 white perch were killed right off the bat. That was the first thing it killed—and a very few bass, a very few suckers, and a very few pickerel, also a few shiners. The white perch was the least resistant of any of the species of fish in that pond.

PRESIDENT TITCOMB: My experience is that the bass is the most resistant of the warm water fishes we have here. The carp is quite sensitive. The northern pike is another very resistant species, much more so than the common pickerel, reticulatus. I wanted to ask Dr. Belding whether in these experiments he had a time limit as to the exposures of these fish in each case?

DR. BELDING: Twenty-four hours—everything is based on twenty-four hours. If you did not set a time limit, your results would be very variable.

PRESIDENT TITCOMB: You find that the effects of the copper sulphate in our largest natural lake, Bantam Lake, a comparatively shallow body of water, the maximum depth being only about twenty-eight feet, the copper sulphate had some action for a period of five days—very little, however, on the last day. I refer here to the clearing up of algae. Observations were taken daily, and it was found that during the first three days it was quite effective. I speak of that because I had got the impression that most of it was precipitated within twenty-four hours.

DR. FIELD: Another element to be taken into consideration in the use of copper sulphate is that the algae floating in the water, such as Anabaena, and so on, will first take up the copper sulphate, thus reducing the amount that reaches the fish and causes it to be in greater dilution than it would be if the water were clear.

PRESIDENT TITCOMB: If it is applied when the first stages of the algae begin to appear, a repetition of the dose may be avoided, and the algae is prevented from getting started so that it becomes very thick, as it does in certain waters of this state.

DR. BELDING: There are three factors that account for all these discrepancies. First, the water; because the conditions in it, such as mentioned by Dr. Field, and the chemical constituents differ, so that you cannot compare one body of water with another. Second, the species of fish vary in their resistance. Third, the method of administering the copper sulphate in the pond, whether it is distributed evenly so that the concentration is uniform and not more pronounced in certain parts of the pond than in others.

DR. DAVIS: In connection with Dr. Belding's results so far as tannic acid is concerned, the Bureau has had a rather interesting experience in one of its hatcheries, apparently due to that cause. At Hartsville, Massachusetts, for a number of years they have had very serious mortality among the brook trout fingerlings in the late winter or early spring. But that happens only in years when there is an excessively heavy rainfall, or when a warm spell comes on following a very heavy snowfall, in other words, when there is a good deal of surface water flowing into the hatchery supply. From further extended inquiry I am satisfied that the mortality among these trout only occurred since the chestnut blight. There was a hillside up above the hatchery which was covered with chestnut trees. Since the chestnut blight attacked the trees of course they have all died and the dead trees have fallen down on the hillside. As you know, chestnut bark is one of the richest in tannic acid, I think, of any of our native trees, and the evidence points rather conclusively to the fact that the tannic acid has got into the water carried down into the hatchery, thus resulting in mortality among the fish. Another interesting point there which coincides with Dr. Belding's experiment is the fact that the rainbow trout were not affected. The brook trout were almost entirely killed in several instances, while the rainbow trout were unaffected—fish of the same age, small fingerlings, still in the sac stage, and others which had been fed only two or three weeks.

DR. BELDING: Brown trout are even more resistant.

DR. DAVIS: There were none at this hatchery.

PRESIDENT TITCOMB: Dr. Stillman, in the survey down there where there was a cleaning out of the trout in one or two streams after the deforestation, was the chestnut a factor there?

DR. STILLMAN: Yes, it is a very common occurrence to find streams which have been devastated apparently affected by the acid from decaying tops. Oak and chestnut are the trees which seem usually to account for this condition, although we see a suggestion of organic pollution from such trees as produce the resins and oils, as, for instance, birch oil, but of course those trees are not so plentiful in lumbering operations. It would be very easy to pick out on a map a dozen streams that are not producing trout at the present time and where the cause seems to be entirely the organic acids from careless lumbering. Regulations and laws should be passed which would compel the lumbermen to clean up their tops after the cutting operations. It seems to affect all the fish life—possibly small pickerel less than other fish; but insect life suffers as well. I think that the blame is often laid on the removal of shade in those streams which fail after lumbering operations, when it is not always that; it is quite as often the organic pollution. Dr. Belding has shown what a small quantity of acid is necessary to poison water, and it must be remembered that when the streams are quite low in the summer we get a greater concentration of the acid in the water than when they are flowing strongly in the winter. I think most of the damage is done in ordinary lumbering under those conditions.

MR. A. H. DINSMORE (Vermont): Several years ago we had some landlocked salmon in a raceway under a steep hillside that had been burned over thirty years before. On the hillside there is now quite a heavy vegetable mould from blueberry bushes and other shrubs that have grown up since the fire. In the summer time, when the weather was intensely warm, we had a cloudburst—I was in the West at the time but these are the facts as they came to me. Almost immediately following that, the salmon, among which we had suffered no loss before, began to die in large numbers. We moved them to other waters, but they all died. I was speaking of that to Dr. Davis on one occasion when he was at our place and he told me of a case where a cloudburst coming in very hot weather produced the same effect. He said it was thought to be due to absorption of oxygen in the water—the surface water running in rapidly and the heat of the ground absorbing the oxygen.

DR. DAVIS: I have a number of instances on record of that sort of thing. Where there are very heavy rains during a hot spell, and there is also a great deal of debris and a large amount of organic matter washed in with the water, the supply of oxygen in the water is practically exhausted, and the resulting mortality has in many instances been laid to that. I do not believe that in any of these cases the cause has been actually demonstrated, but I have known of many cases where that would furnish a logical explanation.

MR. DINSMORE: We made a bulkhead back of that raceway with lumber, thinking we would cut out the surplus water. Last summer we got one of these cloudbursts, and we had in a cement bulkhead, a square cement box that supplies this same raceway, probably four or five hundred trout ranging from four to six inches in length. The water coming into this bulkhead came directly from a pond of about twenty acres. At the time the temperature was high, ranging around 68 to 70 degrees. The trout had made splendid growth all summer, with absolutely no loss. Below that were three or four thousand trout fingerlings, in the raceway where we had lost the salmon—trout that were then from one and a half to two inches long. This shower occurred in the afternoon, and immediately following it, remembering what Dr. Davis had told me, I rushed down to the bulkhead to see what the effect had been on the five hundred fish in the cement bulkhead. I may say that they were not getting any drainage direct from the hillside; they were getting it through a long, narrow neck at the outlet of the pond. The water was rushing down the hillside into this outlet of the pond and then going around into the cement bulkhead. It had been no more than thirty minutes, probably less than that, since the rain started, and there were at least forty dead trout in the bulkhead. Many more were on their backs, and the fish in the raceway below were just beginning to be affected. I jumped into the pond at the head of the cement outlet, dug away some dirt and threw a tremendous flow of the pond water over the surface of the pond, whereupon the fish righted and were all saved. I am sure we would have lost practically all the fish in that bulkhead but for the turning through immediately of the surface waters from the pond.

MR. POWELL: With reference to the chestnut blight, how long would it take the bark of the dead tree to affect the streams?

DR. STILLMAN: We generally see no results the first year. In the cases I have noted personally, the deterioration begins the second year after the cut, and, according to the degree of pollution, it may last possibly six years. In one case I know of an eighteen year pollution, and it is still going on. Ordinarily, though, you can allow twelve to fourteen years to clear.

MR. POWELL: We have had this trouble in Maryland, but although the blight has been on for about seven years, it is just during the past three years that it has been affecting us.

DR. EMBODY: Possibly what I am going to say was covered by Dr. Belding; if it was I beg his pardon. I just want to call attention to two or three conditions resulting from the treatment of a pond with copper sulphate. They have been brought out before; it is nothing new at all, but I think it is well to emphasize them at this time.

First, when you treat a pond with copper sulphate—and you treat it primarily, as I understand, to kill the excessive algae in the pond—you kill not only the algae but also a great many of the food organisms; you kill certain microcrustacea and a great many insects such as mayfly, nymphs, and some others. After these organisms are killed their remains fall to the bottom and begin to decompose, in the process of which they use up the oxygen of the pond. So that you have not only the immediate effects of the copper sulphate but you also have the effects due to lack of oxygen in the pond. The way to overcome that, of course, is, after the treatment with copper sulphate, to run a good supply of water into the pond; you will then overcome that secondary reaction and have only the reaction due to the copper sulphate alone. In killing a large part of the food material or the forage in the pond you are doing something that the fish culturists would not want done, something that the owners of the pond would not want to see brought about in any pond they thought anything of.

MR. DOZE: What experiment would more nearly approach the toxic effect of leaf fall? We have a great leaf fall that kills a great many of our fish. Do you know of any quick remedy for that condition?

DR. BELDING: I do not know exactly what organic acids come from the leaves. I also do not know what the direct action of the acids would be. I do know that in certain of the ponds in Massachusetts in very warm weather, the organic acids and hydrogen-sulphide which are always present to a limited extent in nature, may become detrimental to fish life.