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This study is concerned primarily with the effects of various herbicides and related compounds on the noxious Brazilian water weed, Anacharis(Elodea)densa Planchon, and the spiny-rayed fishes living in association with this plant in some of the coastal lakes of Oregon.

The first attempt made to eradicate this exotic plant was by the Oregon State Game Commission from July 1947 to September 1949. During that period of operations 21 chemicals were tried on Anacharis densa, but only copper sulfate ($CuSO_4.5H_2O$) proved to be partially effective and an economic control. In September 1949 the commission relinquished the weed control project to the Oregon State Agricultural Experiment Station which has since been investigating various methods of controlling the plant. Measures that have been investigated include chemical, mechanical and biological methods of control.

During the spring, summer and fall 1951, extensive screening tests were carried out in the laboratory where 147 different chemicals were tested on Anacharis densa. Of the compounds used, those showing greatest degree of toxicity at the lowest concentration were the copper salts of organic acids and phenolic compounds. Noteworthy of these were copper MCPA, copper 2,4-D, copper phenoxyacetate and copper phenylacetate. Nearly all of the copper salts used would kill the water weed at a concentration of 5 p.p.m. or less in a period of one week. Of the heterocyclic groups pinene and camphene were toxic at fairly low concentrations.

In addition to the laboratory screening tests considerable field work was done at Booth Arm of Siltcoos Lake. Plots were treated with commercial aromatic solvents, "Aromatic #80" and "Socal #3", which effected a 70% and 100% foliage kill respectively. Phenoxyacetic acid was used in the pellet form, but failed to kill the plant at a concentration of 2 pounds to square rod. "Socal #3" when fortified with copper MCPA would make a foliage kill at 4 p.p.m. of the salt and 80 p.p.m. of the solvent.

In conjunction with the water weed screening tests, bioassays were conducted with fish which were present in the coastal lakes in question. Species used were white crappie, Pomoxis annularis Rafinesque; largemouth bass, Micropterus salmoides (Lacepede); and bluegill, Lepomis macrochirus Rafinesque. These tests were conducted so that the degree of toxicity of the chemicals used on the water weed could be determined on the fish. This knowledge is desirable in the event that any of the compounds are used for herbicidal purposes in the coastal lakes of Oregon.

The tests were divided into two series. In the first series a mixed group were used consisting of white crappie, bluegill, and largemouth bass. These tests were conducted in 15 liters of water in a 5-gallon jar. Usually 5 jars constituted a single test. The

duration of each test was 24 hours, and an approximate minimum lethal dosage was ascertained. In the second series the white crappie was used as a test animal and the mortality was determined at 24 and 48 hour intervals. Degree of survival was computed on the basis of a median tolerance limit (TL_m) where 50% of the test fish survived. Test chemicals giving the highest percentage of mortality at the lowest concentrations included 1,2,4-trichlorobenzene, monochlorobenzene, p-chlorotoluene, pentachlorophenate and sodium pentachloroacetate. These compounds were all lethal at 5 p.p.m. or less.

The copper salts of organic acids as well as salts of sodium were not nearly so toxic. The M.L.D. for the copper salts ranged between 30 - 100 p.p.m.; sodium 2,4-D and sodium MCPA had an M.L.D. of 1550 p.p.m. and 300 p.p.m. respectively.

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THE EFFECTS OF HERBICIDES AND RELATED COMPOUNDS ON FISH

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THE EFFECTS OF HERBICIDES AND RELATED COMPOUNDS ON FISH

INTRODUCTION

The following is an account of the studies made on the effects of certain herbicides and related compounds on the Brazilian water weed, Anacharis (Elodea) densa Planchon, and the more common spiny-rayed fishes found in association with this plant in the coastal lakes of Oregon. These bioassays and field observations were conducted as a supplement to the aquatic weed investigation being undertaken by the Oregon Agricultural Experiment Station.

The aquatic weed project having particular reference to the exotic Brazilian water weed in six western Oregon lakes was originally undertaken by the Fisheries Division of the Oregon State Game Commission. Dr. P. R. Needham, formerly director of fisheries for the commission, directed operations at Siltcoos and Tahkenitch Lakes in July 1947 with William Pitney, Aquatic Biologist, in charge of field work. From that time until September 1949, the Game Commission tested 21 chemicals on Anacharis densa; of these only copper sulfate ($CuSO_4 \cdot 5H_2O$) proved to have sufficient herbicidal properties to warrant extensive usage. In September of 1949, the Game Commission relinquished the project to the Oregon State Agricultural Experiment Station which has, in the meantime, been investigating various methods of controlling the water weed. Measures that have been investigated to date include

chemical, mechanical, and biological methods of control.

The study was necessitated by the fact that in recent years two of the most important recreational coastal lakes of Oregon, Siltcoos Lake and Tahkenitch Lake, have become so heavily infested with the weed as to make sports fishing difficult. Boating and logging operations have also been hampered by the presence of this plant. Four other lakes in the Oregon coastal region, Loon Lake, Tenmile Lake, Bradley Lake and Triangle Lake have partial infestations which, unless given thorough periodic treatments, may suffer the same fate as the aforementioned lakes.

The primary purpose of this particular investigation was to find a compound or compounds which, when dispersed among the weeds, would be an effective herbicide and at the same time not be highly toxic to the fishes present. Since the time required for a given compound to kill the weed varies considerably, a tentative testing period of two weeks was set for each laboratory experiment. In actual field work in which experimental plots were treated, observed records were made over varying lengths of time.

In treating fish under laboratory conditions, the test animals were tested for periods of 24 to 48 hours. Actually, however, previous experiments had shown that fish tend to avoid lethal concentrations by moving to a

less toxic area. In other words, the time of treatment set for both the weed and the fish was far in excess of that actually needed for effective treatment.

By and large most of the experimental work was confined to Anacharis densa. From February 1951 until September 1951, over 147 different compounds were screened. Of these the more selective ones were tested on various fishes: white crappie, Pomoxis annularis Rafinesque, largemouth bass, Micropterus salmoides (Lacepede), and bluegill, Lepomis macrochirus Rafinesque, which are representative of the fish fauna of the coastal lakes. Later the white crappie was used exclusively as a test animal, and a median tolerance limit was ascertained at which half the fish survived at 24 and 48 hours respectively. This was achieved by the use of the straight-line graphical interpolation method as outlined by Dr. Peter Douderoff et al (2, p. 1396-1397).

The fish used in these experiments were taken from "potholes" adjacent to the Willamette River. Screening tests were conducted in a laboratory situated on the South Farm near Oregon State College. Field work was carried out principally at Booth Arm of Siltcoos Lake.

All concentrations used throughout the experiments were expressed in parts per million (p.p.m.) which equals one milligram of solute per liter of water.

The work done in this paper is divided into two parts: (1) experimental work done with herbicides and chemicals possessing herbicidal properties on Anacharis densa, and (2) the immediate effects of these compounds on bluegill, white crappie, and largemouth bass. Both tabular and illustrative data covering these experiments is given.

PART I. METHODS AND MATERIALS IN PLANT STUDIES

Collecting and Holding the Water Weed

The Brazilian Water Weed used for laboratory tests was obtained from Siltcoos Lake. The actual collecting technique consisted of taking a small boat out into the lake and then dredging the bottom with a weed-hook. The latter consisted of six pieces of heavy gauge wire embedded in lead inside a 1-inch pipe. Since there was an abundance of the weed during the summer months, it required only a few minutes to fill an ordinary wash tub. During the winter season, however, the plants were considerably shorter in length, and it often required half an hour to obtain a comparable amount.

In collecting the weeds from the lake, care was taken to get the younger and more robust plants, preferably those which were in bloom and with a fair number of buds. This was desirable due to the taxing of the carbohydrate reserve of such a plant when it is growing relatively fast. Hence it is more susceptible to the toxic effects of chemicals and herbicides.

After the weeds were collected, a wet burlap sack was placed over them and they were transported by truck directly to the laboratory at the South Farm and transferred to a large holding tank until such time as they

were needed.

Preparatory to screening tests, sprigs of water weed would be taken from the holding tank and placed in wide-mouthed five gallon jars, figure 4. Each of these jars had previously been filled with 15-liters of water. Chemicals or herbicides were not introduced into the jars until after the temperature of the water equalled that of the laboratory room, and there was evidence of photosynthetic activity.

Illumination

In order to insure adequate illumination, the water weeds being treated were exposed to the radiation of twelve cold cathode fluorescent tubes, figure 4. These were arranged in units of four tubes each. For maximum light distribution one unit was placed directly over the plants, while the remaining two were located on either side of the table supporting the test plants. The period of illumination was set at 12 hours, beginning at 8:00 A.M. and ending at 8:00 P.M. An automatic timing device was installed to perform the duties of turning the lights off and on at the designated time. This device was of considerable value in that the light periodicity of the plant was closely controlled. Any appreciable variation in light duration might have altered the growth pattern of

the plant and the subsequent effects of the test compounds might not have been reliable. (9, p. 386-387)

Temperature

In experimental work of this type as nearly uniform temperatures as possible were desirable and, if possible, an optimum growing temperature range of about 65° - 67° F. should be sustained. If there were any abnormal fluctuations above or below the optimum range, then the toxicity of the chemicals to the plants might be affected because the translocating mechanism of the plant is impaired. (1, p. 223-225)

A relatively stable temperature was maintained in the laboratory room at the South Farm. Since the building was originally of sound construction its heat-holding capacity is noteworthy. During the summer months, the water temperatures remained at about 68° F. with a ± 2 variation. During the colder months of the year, an oil stove was operated continuously, and a temperature of approximately 63° F. was maintained. At all times the temperature was such that photosynthesis was carried on by the plants, and the formation of new buds and shoots was evident.

SCREENING TESTS

The primary purpose in running the screening tests was to determine the minimum concentration at which a compound would kill the water weed. This data was desirable in computing dosages for field operations as it would prove economical to use only the prescribed amount necessary to make an effective kill. The time alloted to each jar under treatment was two weeks, although it was noted that most of the effective compounds made a complete kill in less than one week.

In performing the toxicity tests a random concentration was selected which was believed to be sufficiently toxic to kill the water weed. Following treatment the effects of the chemical were observed on several occasions during the two-week period. At the end of this time the weed was thoroughly examined, and such symptoms as sloughing off of the leaves, softening of the stems, buds, and roots, chlorosis and similar discoloration was recorded. In the event that the weed was not sufficiently affected, and if the chemical being used was believed to hold some promise as a herbicide, then the test was repeated at a slightly higher concentration. Usually in running a duplication, three test jars were used in which the assumed killing concentration was employed in the center jar, and concentrations of lesser and greater

amounts in the other two, the idea being to "pin point" the killing concentration to the minimal amount. If there was any question as to whether or not a plant was completely dead, it would be removed to a jar of fresh water and observed periodically for signs of growth. If the plant were not totally dead, modifications were made in subsequent experiments.

In determining the amount of a chemical to use on the water weed, concentrations were computed in parts per million (p.p.m.). Weighing operations were made with a torsion spring balance accurate to 0.02 grams. In view of the fact that most of the compounds used were of a complex chemical nature, and because of their differential solubilities, it was impossible to standardize them into solutions of known concentrations. Therefore, the concentrations assigned to each test were only fairly accurate, but sufficiently accurate to establish a trend showing a relative toxicity of the compounds used, table 1. Liquid chemicals of known density were not weighed, but were measured out with a pipette.

Chemicals which were not miscible in water, i.e., chlorinated hydrocarbons, benzene homologues, and the heterocyclic groups, were emulsified with special reagents. Several different commercial preparations were used for this purpose and will be discussed in a later

section. Highly insoluble acids were made into water soluble sodium salts before being used on the water weed. For the most part those compounds not containing a 100 per cent ingredient would, if the exact percentage of composition was known, be made up to a concentration equivalent to 100 per cent. This was done to expedite computations.

TABLE 1

THE EFFECTS OF VARIOUS HERBICIDES
ON THE BRAZILIAN WATER WEED, ANACHARIS DENSA.

Effects of toxicity is evaluated as follows: (1) total kill, (2) partial kill and (3) no phytotoxicity is evident.

Compound	Conc. p.p.m.	Cosolvent	Effect
Acetyl diphenylamine	33	acetone, Trex-40	3
Aluminum chloride	200		2
Aluminum chloride and phenoxyacetic acid	10		2
Aluminum nitrate	200		2
p-aminoacetamid and 1% copper chloride	50		2
	20		2
	10		3
Anethole	33		3
Anisic acid	33		1
Aromatic #80	300	5% Tenlo-400	1
	200		1
	100		1
Azoxybenzene plus 1% copper chloride	50		1
	25		1
	10		1
p-benzalaminophenone	30		2
	15		2
	5		3
Benzene hexachloride plus 1% copper chloride	50	acetone and Tenlo-400	2
	25		2
	10		3
Benzophenone plus copper chloride	50	acetone plus Tenlo-400	2
	30		3
	10		3
Benzoyl piperidine	33		3
B-benzoyl propionic acid	66		3
Boric acid	200		1
Camphene plus 1% copper chloride	50	acetone, Tenlo-400	1
	20		1
	10		1
	5		2

TABLE 1--Continued

Compound	Conc. p.p.m.	Cosolvent	Effect
p-chlorobenzene plus 1% copper chloride	50	acetone plus Tenlo-400	2
	20		2
	10		3
p-chlorobenzoic acid	27	acetone	2
3-chloroisopropol-N-phenylcarbamate	50		1
	25		2
	10		3
p-chlorotoluene plus 1% copper chloride	50	Trex-40	2
	20		2
	10		3
Copper acetyl-o-aminobenzoate	10	ammonium hydroxide	1
	5	" "	2
Copper acetyl p-aminobenzoate	10	" "	1
	5		2
Copper acetyl o-aminophenate	10	" "	1
	5		1
Copper acetyl-p-methylaminophenate	10	" "	1
	5		1
Copper 2-aceto-1-naphthate	10	" "	1
	5		2
Copper 2-amino-3, 5-diiodobenzoate	20	" "	1
	10		1
	5		1
Copper o-aminophenol-p-sulfonate	10	" "	1
	5		1
Copper X,4-aminotoluene-3-sulfonate	10	" "	1
	5		1
Copper aniline disulfonate	10	" "	1
	5		1
Copper benzoate	10	" "	1
	5		1
Copper butoxide	10	" "	2
	5		2
Copper d-camphenate	20	" "	1
	10		1
	5		1

TABLE 1--Continued

Compound	Conc. p.p.m.	Cosolvent	Effect
Copper chloride	10		1
	5		1
	2		1
	1		1
	0.5		1
	0.3		1
Copper o-chlorobenzoate	10	ammonium hydroxide	2
	5		3
Copper O-chlorophenoxyacetate	10	" "	1
	5		1
Copper p-chlorophenoxyacetate	20	" "	1
	10		1
	5		2
Copper cyclohexane carboylate	10	" "	2
	5		2
Copper 4, 4'-diaminostilbene	10	" "	1
	5		1
Copper 2,4-dichlorobenzoate	10	" "	1
	5		2
Copper 2,4-dichlorophenoxyacetate	6	triethanolamine	1
	5		1
	4		1
	3		1
	1		2
Copper 2,4-dichlorophenoxyacetate	10	ammonium hydroxide	1
	7		1
	3		2
	1		2
Copper dichloroacetate	20	ammonium hydroxide	1
	10		1
	5		1
Copper 3,5-dichlorobenzoate	10	" "	2
	5		2
Copper 2,4-dihydroxybenzoate	10	" "	2
	5		2
Copper 3,5-dinitrobenzoate	10	" "	1
	5		1
Copper diphenyl-p-p'-disulfonate	10	" "	1
	5		2
Copper ethoxide	10	" "	2
	5		2

TABLE 1--Continued

Compound	Conc. p.p.m.	Cosolvent	Effect
Copper p-flurobenzoate	10	ammonium hydroxide	1
	5		1
Copper hydroxyphenyl	15	" "	2
	7		3
	3		3
Copper idosobenzoate	10	" "	1
	5		1
Copper m-iodobenzoate	10	" "	2
	5		2
Copper p-iodobenzoate	10	" "	1
	5		1
Copper isobutoxide	10	" "	1
	5		1
Copper maleate	10	" "	1
	5		1
	2		1
Copper methoxide	10	" "	1
	5		2
Copper 2-methyl-4-chlorophenoxy acetate	25	" "	1
	10		1
	5		1
	2		2
Copper monochloroacetate	25	" "	1
	15		1
	10		1
	5		2
Copper N-2 chlorophenol phthalmate	10	" "	1
	5		1
Copper alpha-naphthaline acetate	10	" "	1
	5		1
Copper N-1-naphthylphthalate	10	" "	2
	5		2
Copper p-nitrophenylacetate	10	" "	1
	5		2
Copper pentachlorophenate	10	" "	1
	5		1
	2		1
	1		1
Copper phenoxyacetate	40	triethanolamine	1
	30		1
	20		1
	10		1

TABLE 1--Continued

Compound	Conc. p.p.m.	Cosolvent	Effect
Copper phenoxyacetate	5	Triethanolamine	1
Copper phenoxyacetate	10	ammonium hydroxide	1
	7		1
	3		1
	2		2
Copper phenylacetate	10	" "	1
	5		2
Copper succinate	10	" "	2
Copper salt of tannic acid	10	" "	2
	5		2
Copper o-toluate	10		2
	5		3
Copper p-toluate	10	" "	2
	5		2
Copper trichloroacetate	10	" "	1
	5		2
Copper 2,4,6-trinitrophenate	8	" "	1
	5		1
cyclohexane carboxylic acid	33		3
dibenzyl acetic acid	33	acetone	3
9,10-dibromoanthracene	13	acetone and Trex-40	3
4,4'-dibromodiphenyl	25	" "	1
	5		3
2,5-dibromotoluene	33		1
dichloroamine	33	acetone	1
2,4-dichlorobenzonitrile	33	"	3
2,3-dichlorodioxane	33	"	3
3,4-dichloronitrobenzene	15	"	1
	5		1
Diethylbromo-phthalate	33	"	3
2,4-dihydroxybenzoic acid	33	"	3
3,5-diiodo-2-hydroxybenzoic acid	33	"	3
3,5-diiodo-4-hydroxybenzoic acid	33	"	1
2,5-dimethyl furan	33	"	3
1,3-dimethylbenzene-4-sulfonylchloride	33	"	2
2,4-dinitrodiphenylamine	15	acetone and Trex-40	3
	10		3
	5		3
2,4-dinitrophenyl hydrazine	6		1
	14		1

TABLE 1--continued

Compound	Conc. p.p.m.	Cosolvent	Effect
Diphenylamine hydrochloride	30		1
	25		1
	20		1
	15		2
Diphenyl acetic acid	4		2
	3		2
	1		2
Diphenyl carbaminechloride	15		1
	10		1
p-ethoxybenzoic acid	33	acetone	3
ethylene dibenzoate	66	"	3
Ethyl-o-benzoybenzoate	33	"	3
1-ethyloxindole	33	"	3
p-flurobromobenzene	33	"	2
fluarone	33	"	1
furoamide	33	"	3
guanidine nitrate	33	"	3
4,4',4"-hexamethyltriamino-triphenylmethane	30	"	3
	40		3
	20		3
	15		3
N-hexylbenzoate	66	"	3
p-hydroxybenzoic acid	33	"	3
indole	33	"	3
p-iodobenzoic acid	33	"	3
iodosbenzoic acid	33	"	1
isopropyl benzoate	66	"	3
isopropyl-N-phenylcarbamate plus 1% copper chloride	40	Trex-40	2
	25		2
	10		2
K hydrogen-p-sulphobenzoate	33	acetone	3
maleic anyhdride	100		2
O-methoxybenzoic acid	33	"	3
methyl anisate	33	"	2
methyl benzoate	66	"	2
methyl benzoylacetate	33	"	2
methyl-o-benzoylbenzoate	33	"	2
methyl-o-methoxybenzoate	33	"	3
methyl-p-hydroxybenzoate	33	"	3
alpha-naphthelate	30	cellosolvent	2
	10		3
beta-naphthol	33	acetone	1
beta-naphthylamine	33	"	1

TABLE 1--Continued

Compound	Conc. p.p.m.	Cosolvent	Effect
o-nitrobenzoic acid	26	acetone	1
p-nitrobenzoylchloride	33	"	1
phenoxyacetic acid (87%)	25	"	3
87% plus 1% $CuCl_2$	25		2
97%	25		3
96% plus 1% $CuCl_2$	25		2
100% plus 1% $CuCl_2$	25		1
	10		1
phenylbenzoate	66	"	2
o-phenylenediacetamide	33	"	3
o-phenylenediacetic acid	33	"	3
phenylurea	33	"	3
phloroglucinol	33	"	3
N-propul-o-benzoylbenzoate	33	"	2
S-di-M-butylurea	33	"	3
sodium 2,6-dichlorobenzoicindaphenol	33	"	2
p-toluic acid	33	"	3
2,4,6-triaminobenzoic acid hydrochloride	100	"	1
	50		1
	20		2
1,2,4-trichlorobenzene	100	"	1
	50		1
	20		2
3,4,5-trimethoxy benzoic acid	33	"	3
zinc 4-aminotoluen-3-sulfonate	30	ammonium hydroxide	2
	10		1
zinc benzoate	30	" "	1
	10		1
zinc d-campherate	30	" "	1
	10		1
zinc 5-chloro-2-aminobenzoate	30	" "	1
	10		1
zinc p-chlorophenoxy acetate	30	" "	1
	10		1
Zinc 2,4-dichlorobenzoate	10	" "	1
	5		2
	2		2
zinc 2,4-dichlorophenoxyacetate	30	" "	1
	10		1
	5		1

TABLE 1--Continued

Compound	Conc. p.p.m.	Cosolvent	Effect
zinc pentachlorophenate	10	ammonium hydroxide	1
	5		1
zinc phenoxyacetate	30	" "	2
	10		1

FIELD EXPERIMENTS

During the summer of 1951 considerable work was done with herbicides and chemicals on the Brazilian water weed at Siltcoos Lake. The compounds used were, for the most part, selected from among those showing the marked toxicity in the laboratory screening tests. There were, of course, many compounds which showed favorable results, but were not used in field work because of the small quantities on hand.

One of the first compounds to be tested was phenoxy-acetic acid in the pellet form. These pellets had been formulated at the request of Dr. E. J. Kraus of the Horticulture Department, Oregon State College, and manufactured by the Dow Chemical Company. This acid which is normally only slightly soluble in water, had an additive incorporated in some of the pellets which accelerated the solubility considerably. In addition, some of the pellets had 1 per cent copper chloride as a synergist.

During the month of July pellets were tried on thirteen different field plots of plants with varying degrees of success. On the first four trial plots, the pellets were broadcast over a freshly cut area of Anacharis densa at the rate of 2 pounds to the square rod. Periodic checks in August revealed that the acid

in the pellet form was not a very effective herbicide. There was no evidence of a bottom kill, but some of the floating weed showed signs of chlorosis and decomposition. The remaining ten experiments which were tried on growths under different conditions, were no more fruitful so far as an effective kill was concerned. It appeared, then, that the active ingredient in the pellets rose to the surface of the water and made an effective kill there, but failed to do the same with bottom foliage. On table 2 is shown the results of the work done with phenoxyacetic acid on both Anacharis densa and Myriophyllum.

Some measure of success was observed in the use of aromatic solvents and solvents fortified with metallic salts. The aromatic solvent "Socal #3", a product of the Standard Oil Company, with 5 per cent of "Tenlo-400", an emulsifying agent, proved quite effective when applied at the rate of 280 - 330 p.p.m., figure 5. An area was selected in Booth Arm of Siltcoos Lake which was well protected from the prevailing winds, and where the diluting effect of the waves and currents on diffused herbicides was minimized. Plots in this area were treated by means of twin jet nozzles mounted on a 14-foot pole. Effective treatment was attained by spraying the solvent near the root zone. Within three days following the treatment a 100 per cent kill of foliage above the root zone was

realized. Later observations proved that although all the plant tissue above the root zone had been killed, the roots were still viable and young shoots were appearing in the mud.

Experiments were tried using copper salt of 2-methyl-4-chlorophenoxyacetic acid (Cu MCPA) as a synergist with "Socal #3". Since the emulsifying agent, "Tenlo-400", would solubilize the copper salt, there was little difficulty experienced in getting the salt into solution. When this combination was applied at the rate of 4 p.p.m. of the salt to 80 p.p.m. of the solvent, a nearly total kill of the foliage was realized, figure 6. However, this combination required three weeks to complete the kill, whereas "Socal #3" at a considerably higher concentration required only three days. Other copper salts such as copper 2,4-D and copper phenoxyacetate were tried on some of the plots, but due to the difficulty of solubilizing them in the solvent, these experiments never proved satisfactory. In lieu of "Socal #3" plus "Tenlo-400", ammonium hydroxide was used to take copper MCPA into solution. This preparation was applied to a plot at the rate of 10 p.p.m., but failed to make an appreciable kill.

Another commercial solvent, "Aromatic #80", was applied at the same concentration as "Socal #3", but yielded only about a 70 per cent kill. Foliage near the

mud line was not affected, figure 7.

TABLE 2

EXPERIMENTS TESTING PHENOXYACETIC ACID AS POSSIBLE HERBICIDE FOR ANACHARIS DENSA AT SILTCOOS LAKE

Type of Pellet	Remarks	Rate of Application	Location	Result
slow dissolving	1% $CuCl_2$ added	2 lbs/sq. rod	North Beach	negative
slow dissolving		2 lbs/sq. rod	Westlake	negative
fast dissolving	1% $CuCl_2$ added	2 lbs/sq. rod	Westlake	good top kill
fast dissolving	in exposed location	2 lbs/sq. rod	Westlake	negative
slow dissolving	weed cut	2 lbs/sq. rod	Duck Bay	fair top kill
fast dissolving	weed cut 1% $CuCl_2$ added	2 lbs/sq. rod	Duck Bay	good top kill
slow dissolving	weed cut 1% $CuCl_2$ added	2 lbs/sq. rod	Duck Bay	negative
fast dissolving	weed cut	2 lbs/sq. rod	Duck Bay	good top kill
fast and slow dissolving	weed cut, slow with 1% $CuCl_2$ added	2 lbs/sq. rod	Duck Bay	good top kill
fast dissolving		40 p.p.m.	Siltcoos lagoon*	surface kill on Myriophyllum
fast dissolving	1% $CuCl_2$	40 p.p.m.	Siltcoos lagoon*	surface kill on Myriophyllum
slow dissolving		40 p.p.m.	Siltcoos lagoon*	surface kill on Myriophyllum
slow dissolving with $CuCl_2$		40 p.p.m.	Siltcoos lagoon*	surface kill on Myriophyllum

* No Anacharis present.

SUMMARY AND CONCLUSION

In table 1 are listed 147 chemicals used on the Brazilian water weed. Data given show the concentration in p.p.m., the cosolvent needed for solubilizing the compounds, and the effects of the chemicals on the weed expressed numerically. The numerical values ascribed are as follows: (1) this means that the compounds have definite herbicidal effects and did kill the plants at these concentrations, (2) infers that the phytotoxicity is limited, and though the plant shows some of the symptoms common to (1), it is still able to carry on photosynthesis and recover when placed in fresh water, and (3) indicates little or no phytotoxicity.

The chemicals and herbicides listed were, for the most part, obtained from Dr. E. J. Kraus of the Department of Horticulture, Oregon State College. Many of the commercial preparations were procured from Mr. Virgil Freed, Associate Chemist, Agricultural Experiment Station, Oregon State College. All of the copper and zinc salts were prepared at the laboratory on the South Farm. The "Socal #3" had previously been procured by the Oregon State Game Commission while they were in charge of the Aquatic Weed Control project at Siltcoos Lake.

In general the compounds giving the greatest percentage of kill were the copper salts. Most noteworthy

of these were the salts of 2,4-D, 2-methyl-4-chlorophenoxyacetic acid and phenoxyacetic acid. Under laboratory conditions these compounds would make an effective kill in less than a week at 1-2 p.p.m.

When used as a synergist with an aromatic solvent, Copper MCPA increased the toxicity 400 per cent over that of the unfortified solvent. The copper salts of organic acids were found to be more effective herbicides when considering the percentage of copper present in the molecule than in inorganic copper salts. This is of considerable importance from a biological point of view, because when, for example, copper sulfate is used as a herbicide it deposits a residue of the metal on the lake bottom which acts as a sterilent to bottom fauna for an indefinite period. (8, pp. 83-84)

Organic copper salts have their limitations because of their high degree of insolubility. In water there is no evidence of their going into solution. Triethanolamine will take them into solution at the rate of 200 grams of solvent to 1 gram of the salt. Unfortunately this material is comparatively expensive and its use would be prohibitive on a wholesale basis. Ammonium hydroxide (28%) will solubilize the salts at the rate of 6:1. However, in using this as a solvent, the copper complex resulting is not one of a simple copper cation and an

anion of the acid radical, but a complex of dissimilar groups whose exact chemical properties are unknown. (4, p. 161) This should warrant further investigation.

Zinc salts proved highly satisfactory at low concentrations, but due to limitations of time and material these were not fully exploited in the field and laboratory. Further investigation is required to realize their full herbicidal value.

Inorganic copper salts were effective contact herbicides, but failed to make a root kill. When the foliage was exposed to a concentration as low as 0.3 p.p.m. of copper chloride, the tissues would commence to decompose within 10 days. A concentration of 5 p.p.m. would kill the weeds in 4 days. Copper sulfate in the crystal form has been used for several years on this water weed and will destroy it at about 5 p.p.m. The copper salt, as mentioned previously, is not desirable because of the sterilizing effects it has on bottom fauna.

Pentachlorophenol and its copper and sodium salts proved excellent results in the laboratory, but due to their inherent toxicity to aquatic vertebrates, their general application is not encouraged. (6, p. 869) Tests show that this compound and its homologues will kill white crappie at a concentration as low as 0.18 p.p.m.

Figure 1. Section of laboratory where solutions were prepared for bioassay determinations.

Figure 2. Holding tank where fish were held before being used in toxicity tests.

Figure 3. Water-baths and test jars used in conducting toxicity tests on fish.

Figure 4. Anacharis densa undergoing treatment with different herbicidal materials.

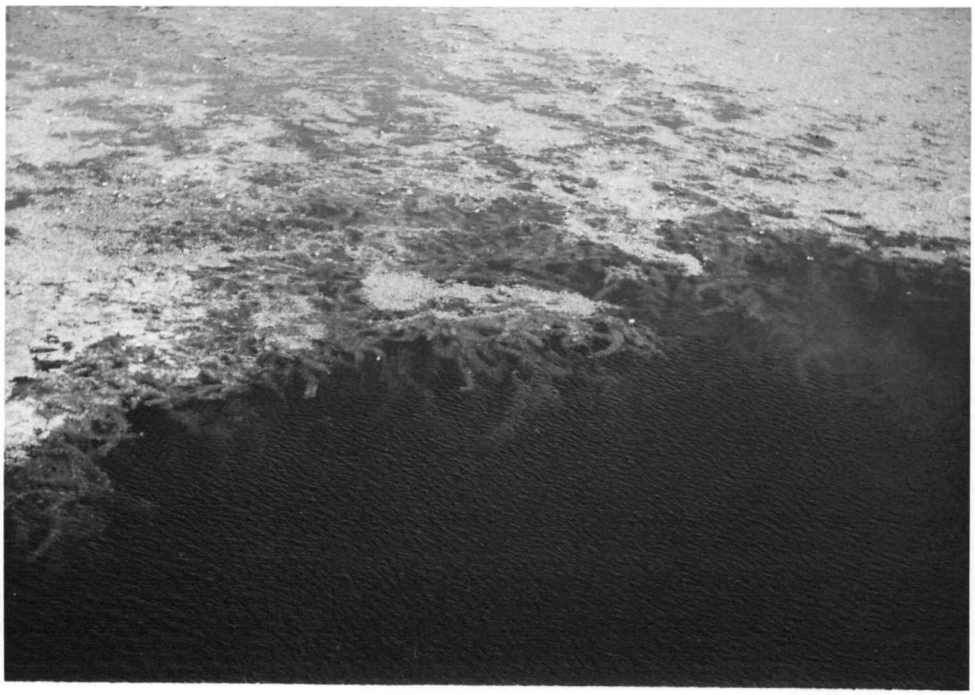


Figure 5. Anacharis densa following treatment with 300 p.p.m. of "Socal #3". Foreground shows treated area.

Figure 6. Anacharis densa following treatment with 4 p.p.m. of copper MCPA and 80 p.p.m. of "Socal #3". Nearly a 100% kill was realized.

Figure 7. Anacharis densa after treatment with "Aromatic 80". Only a 70% kill was effected.

Figure 8. Anacharis densa treated with 10 p.p.m. of copper MCPA and 40 p.p.m. of "Socal #3". Picture taken two weeks following treatment.

PART II. METHODS AND MATERIALS USED IN FISH STUDIES

Collecting Test Fishes

In the early stages of the toxicity experiments the test animals used were principally the three-spined stickleback, Gasterosteus aculeatus Linnaeus, and the redsided shiner, Richardsonius balteatus (Cope). It was soon learned, however, that the redsided shiner required relatively higher amounts of dissolved oxygen than most fish, and hence could not be transported readily in the 5-gallon collecting jars. The stickleback, on the other hand, could easily be transported, but could not survive for long in the city tap water which was used for these experiments. Owing to these inadequacies, the use of these two fishes was discontinued.

Since this problem deals primarily with the fishes of the coastal lakes, the obvious test animals to use were the Centrarchids (bass family), cutthroat trout and silver salmon. Of this group of fish the Centrarchids were found to be the easiest to handle, and suffered the smallest amount of mortality while being transported in the carrying jars. The trout and salmon, though a very important species with respect to this particular problem, were collected on several occasions, but could never be held for a period of sufficient duration for bioassays.

In nearly every incidence these fish died within a 24 hour period. The causative factor responsible for such a mortality was believed to be zinc which leached out of the water pipes.

During the summer of 1951 most of the fish were obtained from several large potholes located on the land of C. E. Hahn, 8 miles south of Corvallis. During this period several hundred bluegill, white crappie and largemouth bass were taken. In the fall of 1951 a relatively long pothole was discovered two miles west of the Hahn place, which yielded approximately 2,000 white crappies. Nearly all the fish taken from this pothole were of a uniform size and age group. With this fish stock it was possible to carry on experiments using one species of fish exclusively. Heretofore, experiments had been confined to mixed species groups because of the shortage of fish.

The equipment employed in seining test fish consisted of three different seines and several 5-gallon glass jars. In seining a pothole over 50 feet wide and deeper than 3 feet, a 50 by 6 foot seine having a one-half inch mesh was used. For smaller bodies of water, either a 25 by 5 foot, or 10 by 4 foot seine would be used. No elaborate technique was used in taking the fish with a seine other than that care was taken in selecting a landing site having a gradual

slope. This was desirable as it helped in avoiding loss or injury to an unnecessary number of fish.

Preparatory to seining operations several 5-gallon carrying jars were filled with clean water. This was necessary because once seining commenced in the confines of a small pothole, there was a tendency to rile the water from the bottom mud. After the seine was drawn to the landing site, it would be "pursed" in order to prevent any fish from escaping. This was done by drawing the lead line in close to shore, and leaving the cork line extended out a few feet. After the fish were pocketed they were carefully removed to the 5-gallon jars. In transferring the fish from the net to the collecting jars, only those species which were to be used in the tests were retained. Trash fish, such as suckers, squaw fish, and carp, which were believed to be competing for food with the test fish, were killed and thrown on the land.

The number of fish placed in a jar was determined by their sizes, the water temperatures, and the time required to transport them to the holding tank. (3, p. 105). Usually no more than 15 fish having an approximate average length of 11 centimeters would be taken in a single jar. If, for example, the collected fish had to be transported on a hot day, the water in the jars would be changed once or twice at different points while en route to the laboratory. This was done to insure adequate aeration, and to

keep the water temperature down.

Holding Fish

For the purpose of holding fish until such time as they were needed in the bioassays, an 8 1/3 x 2 3/4 x 2 1/2 foot tank was used, figure 2. The tank was divided into three separate compartments, each of which had an overflow chamber four inches below the top of the main tank. When this tank was first used a large number of fish were killed due to the leaching out of toxic materials from the zinc chromate paint which was used as a preservative. This condition was corrected by painting the tank with B-asphaltum, a non-toxic and water insoluble preparation.

To maintain adequate aeration, water was piped to the holding tank from the laboratory and directed into each compartment by a spray nozzle. The jet-like action caused the surface of the water to circulate and consequently kept it replenished with about 9 p.p.m. of dissolved oxygen. According to Welsh any induced surface action such as is brought about by waves, wind, or other agencies will frequently keep surface waters at a condition approaching saturation. Physically, surface action, whether caused naturally or artificially, should be similar.

The water used in the holding tank and in the laboratory experiments proved satisfactory at all times. The residual chlorine was never sufficiently high to be toxic to the fish. The hydrogen-ion concentration was nearly always alkaline, and only rarely exceeded pH 6.7. Total alkalinity, which was checked periodically, never exceeded 60 p.p.m. as calcium carbonate. Since fresh water was continuously being fed into the tank, the water temperature was kept fairly low (63° F.) during the summer months; and never froze over during the winter months.

During the holding period fish were fed every other day. Their diet consisted mainly of ground liver and aquatic insect larvae of various kinds. (2, p. 1393) The liver was ground with an ordinary kitchen meat grinder and then dispersed in the water by agitating it with a glass rod. Liver which was not eaten would be removed from the tank bottom with a rubber siphon hose. This was done to prevent bacterial or fungus infestations. An attempt was made to culture Daphnia sp. as a food organism, but in view of the high protein requirements of a fairly large number of fish (there were normally over 150 fish on hand at one time), this plan did not prove feasible.

Periodically the holding tanks would be drained and thoroughly cleaned; if necessary an additional coating of

B-asphaltum would be applied. This was necessary because of the large amounts of metabolic wastes that would accumulate as well as bits of liver which were missed by the siphon hose. Throughout the duration of the experimental work there was no appreciable loss of fish in these tanks. All fish which died in the tanks were immediately removed.

Test Jars and Temperature Control

All of the fish toxicity tests were carried out in 5-gallon (19 liters) jars, figure 3. The jars were of the wide-mouth type and cylindrical in design. These features were important when considering that some of the toxic substances used would cause the fish to swim violently about. Had a rectangular aquaria been used, the test animals doubtless would have injured themselves by swimming against the corners. With the cylindrical jars fish were compelled to swim in a circular pattern, thus avoiding injury.

Truly uniform water temperatures were maintained by keeping the jars immersed in water baths. There were, in all, four of these water baths made of 24-gauge galvanized sheet metal. Each unit was 10" x 16" x 6.5 feet and would accommodate 6 test jars, figure 3. During the summer and fall months, the temperature was relatively constant due

to the heat-retaining properties of the laboratory. The mean temperature at this time was 63° F. ± 3°. During the colder months of the year thermostatically controlled heating coils were used in two of the units. However, these were seldom used because the laboratory could be kept at mean temperature of 60.5° F. ± 2° by use of an oil stove which operated continuously.

Selecting and Acclimatizing Test Fish

Fish to be used for the toxicity tests were first carefully inspected for signs of fungus infection, bruises, and other abnormalities which might impair their usefulness as test animals. Fish of questionable fitness were never used. Fish of nearly uniform size were selected. To have used a group of fish having a considerable difference in length would probably have made tests invalid because of the fact that smaller fish in some species may be more susceptible to some toxic substances than are the larger ones. The length of the largest fish used should not be more than 1.5 times as long as the smallest specimen used. (2, p. 1383)

TEST CHEMICALS

The compounds selected for use on the test fish were, for the most part, herbicidal in nature. Many of the toxic materials used were commercial herbicides such as sodium 2,4-D, table 16; IPC (Isopropyl-N-phenylcarbamate), table 12; sodium MCPA (sodium 2-methyl-4-chlorophenoxyacetate), table 17; and TCA (trichloroacetic acid), table 40. Other preparations which had not as yet been released to the public were tested. An example of this was CMU or p-chlorophenyldimethylurea, table 24. Aromatic solvents commonly used on aquatic weeds were tested. Among these were "Aromatic #80", table 4, and "Socal #3", table 5. Some compounds showing herbicidal properties, but having no commercial significance were used in a number of bioassays. Among these were homologues of the commercially proven herbicides, i.e., phenoxyacetic acid, figure 5, and p-chlorophenoxyacetic acid. Several chlorinated and nitrated compounds were used. Aromatic compounds were tried because of the phytotoxic effects exerted by them on the Brazilian water weed. Heterocyclic groups were found to be quite toxic to the test animals as well as the water weed. Chemicals belonging to this group included pinene, figure 52; camphene, table 18; and turpentine, table 23. Of the inorganic salts used, the metallic ones showed the

greatest toxicity to the fish. Foremost among these were copper chloride, table 21, and copper sulfate, table 43.

Since so many of the compounds used were water insoluble, it was necessary to solubilize, or emulsify them with special reagents. As these substances were to be exposed to the test fishes also, it was deemed necessary to determine the extent of their toxicity so as to ascertain whether or not the reagent or the test chemical affected the test animal most. Solubilizing reagents showing little or no toxicity at relatively high concentrations (over 300 p.p.m.) included acetone, butylcellosolve, ethanol, triethanolamine and "Tween-20", table 3. Reagents having a marked toxicity at lower concentrations (25 p.p.m. or less) were primarily the commercial emulsifiers. Those tested on fish included "Tenlo-400" and "Trex-40", table 19.

PRELIMINARY BIOASSAYS

With few exceptions nearly all compounds used in these tests had not previously been used in bioassay work. It was therefore necessary to run preliminary tests in order to conserve fish. The procedure employed was the same as was used in the final experiments. In the tests using mixed groups, i.e., white crappie, bluegill, and largemouth bass, one fish of each species would be placed in a volume of 10 liters. The concentration used was strictly a trial and error proposition. Obviously a considerable number of fish were lost before a minimum lethal dosage (M.L.D.) was arrived at. The M.L.D. is the minimum concentration of toxic material necessary to kill all of the test fish. In prognosticating the approximate concentration of a given chemical, it was often useful to note the minimum lethal dosage of one of its homologues, and use that as an initial concentration. For example, it was found the 0.56 p.p.m. of sodium pentachloroacetate would constitute the M.L.D. for white crappies. Using this as an index it was fairly easy to arrive at a concentration of sodium pentachlorophenate needed to produce an M.L.D. on white crappies. In this particular case the dosage was 0.18 p.p.m. at a temperature of 62.3° F.

In running preliminary screening tests with white crappies as the exclusive test animal, reference was

always made to tests previously conducted on the mixed groups. This was of considerable assistance in "pin pointing" the required concentrations.

The toxicity tests were divided into two separate series: in the first series the white crappies, bluegills, and largemouth bass were used, while in the second series the white crappies were used as an exclusive test animal. In the first series the specific forms were usually arranged differently in each of the test jars, i.e., in the first test jar there might be 2 bluegills, 2 largemouth bass, and 1 white crappie, while in the next test jar this procedure would be reversed. The purpose for doing this was to offset the effects of a high mortality on just one species if it were particularly susceptible to a given compound.

The test fish were exposed to the toxic materials for a period of 24 hours. Perhaps a longer exposure would have given more pertinent data, but as previously mentioned the purpose for conducting these tests was to find out the general effects of herbicidal materials on fish. Since any poisoning operations on the coastal lakes of Oregon would prove both slow and costly, the process would at best treat only small segments of the lake at a given time; this should allow any fish in an area being treated to swim from a concentrated to a less

concentrated area, and this doubtless could be done in less than 24 hours.

Laboratory tests showed that white crappies were more susceptible to the toxic materials than were the largemouth bass, while the bluegills were the least effected. Results of this series of experiments are presented in tables 4 to 20.

With the second series of bioassays only the white crappie was used as a test animal. This procedure was followed so that the more significant compounds which had previously been used on the mixed groups of fish could be tested. Douderoff et al suggested that at least 10 test animals be used to an experiment, but as in the case of the mixed groups, the number of test fish were limited to 5 per jar because of the large number of chemicals which was used on this fish. (2, p. 1395) Doubtless had 10 fish been used to an experiment the results would have been more significant. However, since only 5 test animals were used, extreme care was exercised in conducting the tests, and if the tests tended to be erratic they would be repeated. Any irregularity in survival was believed to be caused by some inherent defect in one or more of the test animals. For example, while making a bioassay with 2,4-dinitrophenol the results of the first two series (this involved 5 test jars and 25 fish in each series) were not

consistent with respect to the concentration of the material used. In the first test there were more fish killed by an intermediate concentration than by a higher one. In the second test, which was an exact replica of the first except for the test animals, fish died in the lesser and greater concentration, but survived in the intermediate ones. With the third test, and again this was a replica of the first, fish mortality was observed to bear a direct relationship with respect to the concentration of toxic materials used.

CONDUCTING TOXICITY TESTS

Preparatory to running the bioassays, all test containers were filled with tap water to a volume of 15 liters. The water temperature usually was nearly equal to that of the holding tank. This condition was helpful in that it was not always necessary to temper the water before proceeding with an experiment. Five fish were placed in each test jar and left there for a period of 2 to 3 days. A minimum of 2 days was required for acclimatizing the fish to the new environment. At the end of this period the test fish were re-examined, and if there was evidence of distress or any abnormalities noted the questionable specimens were removed.

Throughout the holding period all jars were aerated. The source of aeration was a 1/8 horse power motor which activated a small pump. Attached to this was a rubber hose having a diameter of about 1/8 inches. Branching off this and leading to each of the test jars was a similar hose with an air diffuser connected to the open end. This pump could supply ample air for 24 jars.

Since the approximate concentration had been determined in the preliminary tests, the compound to be tested would be first solubilized in water from one of the test jars. It was then poured into a jar and thoroughly diffused with a glass rod. At this time the pH and the

temperature of the water was recorded. When volatile compounds were used the solutions were not aerated and the dissolved oxygen would be determined. However, in the case of metallic salts and other non-volatile compounds the water would be aerated by means previously explained. This was necessary because of the precipitation of the mucosa on the fish's gills caused by most of these salts. (3, p. 74) Since these tests were not conducted primarily out of detached academic interest, but rather to simulate conditions as they might be if poisoning operations were conducted on the coastal lakes of Oregon, then the aeration procedure seemed necessary. Both the aerated water in the jars and water tested at Siltcoos Lake held between 8-10 p.p.m. of dissolved oxygen. To what extent the aeration affected the initial toxicity of the chemicals is not known. There is some evidence that work done by others closely parallels that done on the white crappies. Goodnight found that 0.2 to 0.6 p.p.m. of sodium pentachlorophenate was lethal to fresh-water fishes in non-aerated water. (5, p. 869) Using aerated water at a concentration of 0.18 p.p.m. of the same compound proved lethal to the white crappies.

The M.L.D. of copper sulphate for fresh-water fish in non-aerated water has been reported as ranging from 0.3 to 10 p.p.m. with the lesser concentrations being effective

in acid waters. (3, p. 74) In aerated water having a pH 7.1, the M.L.D. for white crappies was 2.4 p.p.m.

All of the copper salts, which includes derivatives of phenolic compounds and organic acids, acted similarly in their behavior as a water weed herbicide and, within a reasonably narrow range, did not vary appreciably in their toxicity towards fish. Those copper compounds which were least toxic to fish and produced an M.L.D. somewhere between 30 p.p.m. and 100 p.p.m. include copper monochloroacetate, copper p-nitrophenyl acetate, copper MCPA, copper phenylacetate, copper p-chlorophenoxyacetate, copper alpha-naphthaleneacetate, copper phenylacetate, and copper o-toluate.

Copper salts which produced an M.L.D. at concentrations less than 30 p.p.m. includes copper dichloroacetate, copper d-camphenate, copper benzelate, and copper 2,4-D.

Salts of the lighter metals showed the least toxicity towards fish of any of the materials used. Sodium 2,4-D effected a 100 per cent kill at 1550 p.p.m., while Sodium MCPA produced similar results at 30 p.p.m.

Chlorinated aromatic compounds were extremely toxic, and produced an M.L.D. at very small concentrations. 1,2,4-trichlorobenzene inflicted a total loss of fish at 5 p.p.m.; monochlorobenzene at 2 p.p.m. and p-chlorotoluene

was lethal at 5 p.p.m.

Emulsifying reagents were, for the most part, extremely toxic to fish, and are discussed more in detail in the conclusion.

Commercial aromatic solvents were comparable in their effects to the chlorinated aromatic compounds. "Socal #3" produced an M.L.D. some place in excess of 10 p.p.m. "Aromatic #80" killed all of the test fish at 10 p.p.m.

When the white crappie was used as an exclusive test animal the effects of various compounds upon this fish varied appreciably in its toxicity than when used on the mixed groups of fish. For instance, Sodium MCPA effected an M.L.D. on the mixed groups at 300 p.p.m., while with the white crappie only 180 p.p.m. was required. Only 2.40 p.p.m. of 1,2,4-trichlorobenzene was needed to kill white crappie, and 5 p.p.m. for the mixed groups. When using copper dichloroacetate, however, 18 p.p.m. were needed to inflict a total loss on white crappie, while with the mixed groups somewhere between 10-30 p.p.m. were required. This would infer, then, that there is a mutual tolerance towards copper dichloroacetate, but with the other compounds the white crappies are more susceptible at lower concentrations. Mutual tolerance was also shown for the following compounds: monochlorobenzene, "Socal #3" and pinene.

TABLE 3

TOXICITY OF CERTAIN COSOLVENTS AND EMULSIFIERS TO FISH

Chemical	Concentration (p.p.m.)	Duration (hours)	No. Fish	Per cent of Mortality
Ammonium hydroxide	5	48	5	100
Acetone	500	48	6	0
Butyl cello-solve	1000	96	5	0
Dioxane	400	48	5	0
Ethanol	1000	96	5	0
Methanol	700	48	7	0
"Tenlo-400"	4	24	6	100
Triethanolamine	1000	48	6	0
"Tween-20"	300	48	6	0

TABLE 4

THE EFFECTS OF "AROMATIC #80" - 5% "TENLO-400"
ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	8.4	8.2	9.5	8.3	0
1	5	8.4	8.2	9.5	8.4	0
2	5	8.5	8.2	9.4	8.4	0
5	5	8.5	8.3	9.4	8.2	0
10	5	8.5	8.4	9.4	8.4	100

TABLE 5

THE EFFECTS OF "SOCAL #3" - 5% "TENLO-400"
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liter jars at a temperature of 63° F.)

Concentration in p.p.m.	Number Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	8.2	7.9	9.4	7.4	0
1	5	8.2	8.0	9.4	7.0	0
2	5	8.2	8.0	9.3	7.1	0
5	5	8.0	7.8	9.4	6.8	40
10	5	8.0	7.8	9.3	6.5	40

TABLE 6

THE EFFECTS OF COPPER DICHLOROACETATE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 64° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
2	5	8.2	8.0	Aerated	Aerated	0
5	5	8.2	8.0			0
10	5	8.2	7.8			40
30	5	8.6	8.3			100
50	5	9.2	8.6			100

TABLE 7

THE EFFECTS OF 2,4-DINITRODIPHENYLAMINE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 64° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	8.4	8.2	9.4	9.2	0
15	5	8.5	8.1	9.4	9.3	0
30	5	8.7	8.3	9.3	9.2	0
50	5	9.0	8.8	9.4	9.0	20
75	5	9.3	8.9	9.4	9.0	0

TABLE 8

THE EFFECTS OF 9,10-DIBROMOANTHRACENE
ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
1	5	7.8	7.5	9.6	9.5	0
3	5	7.8	7.4	9.6	9.4	0
5	5	7.8	7.4	9.6	9.4	0
10	5	8.0	7.6	9.6	9.3	40
25	5	8.0	7.7	9.6	9.3	100

TABLE 9

THE EFFECTS OF COPPER MONOCHLOROACETATE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liter jars at the temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	8.2	8.0	Aerated	Aerated	0
15	5	8.2	8.0			0
25	5	8.6	8.3			0
50	5	8.8	8.4			60
100	5	9.0	8.3			80

TABLE 10

THE EFFECTS OF P-CHLOROTOLUENE
ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	8.2	8.0	9.4	8.6	20
1	5	8.2	8.0	9.4	8.5	60
5	5	8.2	8.1	9.4	8.3	100
10	5	8.3	8.0	9.3	8.2	100
15	5	8.3	8.0	9.3	8.2	100

TABLE 11

THE EFFECTS OF 3,4-DINITROBENZENE
ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	7.8	7.6	9.8	9.0	0
1	5	7.8	7.8	9.8	9.2	0
5	5	7.9	7.7	9.8	9.1	40
15	5	8.2	7.8	9.6	8.9	100
25	5	8.4	8.0	9.5	8.8	100

TABLE 12

THE EFFECTS OF ISOPROPYL-N-PHENYLCARBAMATE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
1	5	8.2	7.9	9.4	9.4	20
5	5	8.2	8.0	9.4	9.3	40
15	5	8.3	8.0	9.5	9.4	100
30	5	8.5	8.1	9.5	9.4	100
40	5	8.8	8.3	9.5	9.4	100

TABLE 13

THE EFFECTS OF DIPHENYLAMINE HYDROCHLORIDE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 59° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	7.4	7.0	10.1	9.9	0
1	5	7.4	7.0	10.0	9.8	0
5	5	7.4	6.9	10.0	9.8	80
10	5	7.4	6.8	9.8	9.7	100
15	5	7.4	6.9	9.8	9.7	100

TABLE 14

THE EFFECTS OF 1,2,4-TRICHLOROBENZENE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	7.8	7.6	9.5	9.5	0
1	5	8.0	7.6	9.5	9.4	0
3	5	8.0	7.8	9.5	9.5	80
5	5	8.2	7.9	9.4	9.3	100
10	5	8.4	8.0	9.4	9.3	100

TABLE 15

THE EFFECTS OF MONOCHLOROBENZENE
ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	8.0	7.8	9.6	9.6	40
2	5	8.0	7.7	9.6	9.5	100
5	5	8.0	7.8	9.5	9.5	100
15	5	8.4	8.0	9.5	9.4	100
25	5	8.7	8.1	9.5	9.4	100

TABLE 16

THE EFFECTS OF SODIUM 2,4-DICHLOROPHENOXYACETATE ON WHITE CRAPPIE.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
750	5			9.6	9.4	0
870	5			9.6	9.3	0
1150	5			9.6	9.4	0
1350	5			9.6	9.2	40
1550	5			9.6	9.2	100

TABLE 17

THE EFFECTS OF SODIUM 2-METHYL-4-CHLOROPHENOXYACETATE ON WHITE CRAPPIE, BLUEGILL, AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
10	5	7.6	7.3	9.5	9.5	0
35	5	7.6	7.2	9.5	9.4	20
75	5	7.6	7.2	9.5	9.4	60
150	5	7.8	7.3	9.4	9.2	40
300	5	8.2	7.2	9.4	9.3	100

TABLE 18

THE EFFECTS OF CAMPHENE ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 59°F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
0.5	5	7.5	7.2	10.0	9.7	20
2	5	7.5	7.2	10.2	9.8	60
5	5	7.5	7.0	10.2	9.8	100
10	5	7.5	7.0	10.0	9.8	100
25	5	7.5	6.9	10.0	9.8	100

TABLE 19

THE EFFECTS OF "TREX-40" ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 59° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
2	5	8.2	7.8	9.8	9.8	20
3	5	8.2	7.9	9.8	9.7	20
5	5	8.2	7.9	9.8	9.7	80
25	5	8.8	8.3	9.8	9.7	100
50	5	9.0	8.7	9.8	9.7	100

TABLE 20

THE EFFECTS OF PINENE ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 61°F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
1	5	7.5	7.0	9.7	9.5	0
3	5	7.5	7.0	9.8	9.5	60
5	5	7.5	6.9	9.7	9.6	80
10	5	7.5	6.9	9.7	9.5	100
20	5	7.5	6.8	9.7	9.3	100

TABLE 21

THE EFFECTS OF COPPER CHLORIDE ON LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 59° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
3	5	7.6	7.3	Aerated	Aerated	80
5	5	7.6	7.3			80
7	5	7.6	7.2			100
10	5	7.6	7.2			100
15	5	7.6	7.0			100

TABLE 22

THE EFFECTS OF DIPHENYLACETIC ACID
ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	7.8	7.6	9.7	9.5	0
10	5	7.8	7.2	9.7	9.4	20
20	5	7.8	7.2	9.7	9.4	60
30	5	7.6	7.0	9.6	9.4	100
50	5	7.6	6.9	9.5	9.2	100

TABLE 23

THE EFFECTS OF TURPENTINE ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
10	5	7.4	7.1	9.6	9.5	80
20	5	7.4	7.1	9.6	9.4	100
50	5	7.4	6.9	9.5	9.5	100
100	5	7.4	6.9	9.5	9.5	100
200	5	7.4	6.8	9.4	9.3	100

TABLE 24

THE EFFECTS OF P-CHLOROPHENYL DIMENTHYLUREA-"CMU", ON BLUEGILLS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	7.4	7.1	Aerated	Aerated	0
10	5	7.4	7.0			0
30	5	7.4	6.8			20
50	5	7.4	6.7			0
100	5	7.3	6.5			80

TABLE 25

THE EFFECTS OF COPPER P-NITROPHENYLACETATE ON WHITE CRAPPIE AND BLUEGILL.
(Tests conducted were in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
10	5	8.6	8.4	Aerated	Aerated	0
20	5	8.8	8.4			20
30	5	8.9	8.6			60
50	5	9.1	8.7			100
100	5	9.3	9.1			100

TABLE 26

THE EFFECTS OF COPPER D-CAMPHENATE ON WHITE CRAPPIE AND BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
2	4	8.7	8.5	Aerated	Aerated	0
5	4	8.7	8.4			0
10	4	8.8	8.5			25
15	4	8.9	8.6			50
20	4	9.2	8.9			100

TABLE 27

THE EFFECTS OF COPPER BENZILATE ON BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
2	5	8.8	8.4	Aerated	Aerated	0
5	5	8.8	8.3			0
10	5	8.8	8.2			0
15	5	8.9	8.5			20
20	5	9.3	9.0			80

TABLE 28

THE EFFECTS OF COPPER 2-METHYL-4-CHLOROPHENOXYACETATE ON WHITE CRAPPIE, BLUEGILL AND LARGEMOUTH BASS.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
2	5	8.4	8.0	Aerated	Aerated	0
5	5	8.4	8.1			20
10	5	8.5	8.2			20
20	5	8.7	8.2			20
30	5	9.0	8.4			60

TABLE 29

THE EFFECTS OF COPPER PHENOXYACETATE ON WHITE CRAPPIE AND BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
2	5	8.2	7.8	Aerated	Aerated	0
5	5	8.2	7.9			40
10	5	8.2	7.9			20
20	5	8.4	8.1			20
30	5	9.1	8.5			60

TABLE 30

THE EFFECTS OF COPPER P-CHLOROPHENOXYACETATE ON BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	8.7	6.6	Aerated	Aerated	0
10	5	8.8	7.0			0
20	5	8.9	7.6			20
30	5	9.0	8.1			0
50	5	9.2	8.4			60

TABLE 31

THE EFFECTS OF COPPER ALPHA-NAPHTHALENEACETATE ON BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	8.6	6.3	Aerated	Aerated	0
10	5	8.6	6.7			0
30	5	8.8	7.3			0
50	5	10.2	9.0			40
75	5	10.4	9.5			100

TABLE 32

THE EFFECTS OF COPPER PHENYLACETATE ON WHITE CRAPPIE AND BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
10	5	8.3	6.5	Aerated	Aerated	0
20	5	8.3	7.0			0
30	5	8.6	7.8			20
50	5	9.4	8.3			40
75	5	9.5	8.4			100

TABLE 33

THE EFFECTS OF COPPER O-TOLUATE ON BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
10	5	8.2	6.5	Aerated	Aerated	0
20	5	8.4	6.7			0
30	5	8.7	7.6			20
50	5	9.5	8.6			60
75	5	9.6	8.8			100

TABLE 34

THE EFFECTS OF COPPER 2,4-DICHLOROBENZOATE ON WHITE CRAPPIE AND BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	8.4	8.2	Aerated	Aerated	0
10	5	8.4	8.1			0
20	5	8.7	8.5			40
30	5	9.0	8.7			100
50	5	9.0	8.6			100

TABLE 35

THE EFFECTS OF ZINC 2,4-DICHLOROPHENOXYACETATE ON BLUEGILL.
(Tests were conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in 48 hours
		Start	End	Start	End	
5	5	8.5	8.3	Aerated	Aerated	0
10	5	8.6	8.4			0
20	5	8.9	8.7			20
30	5	9.2	8.7			100
50	5	9.3	8.9			100

TABLE 36

THE EFFECTS OF SODIUM 2,4-DINITROPHENATE ON WHITE CRAPPIE.
(Tests were conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
5.6	5	7.7	7.4	9.8	9.6	0	0
7.5	5	7.7	7.5	9.6	9.3	0	20
10.0	5	7.6	7.5	9.7	9.4	40	80
13.5	5	7.8	7.4	9.6	9.3	80	100
18.0	5	7.7	7.4	9.6	9.3	100	100

TABLE 37

THE EFFECTS OF SODIUM 2-METHYL-4-CHLOROPHENOXYACETATE ON WHITE CRAPPIE.
(Tests conducted were in 15 liters of solution at a temperature of 59° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
36	5	7.3	7.2	9.7	9.5	0	0
56	5	7.3	7.2	9.7	9.3	0	0
75	5	7.4	7.5	9.4	9.2	60	40
135	5	7.7	7.3	9.4	9.0	80	80
180	5	7.7	7.4	9.4	9.1	80	100

TABLE 38

THE EFFECTS OF SODIUM PENTACHLOROACETATE ON WHITE CRAPPIE.
(Tests conducted were in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
0.18	5	7.3		Aerated	Aerated	0	20
0.24	5	7.3				0	40
0.32	5	7.3				40	40
0.42	5	7.3				60	80
0.56	5	7.3				100	100

TABLE 39

THE EFFECTS OF SODIUM PENTACHLOROPHENATE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
0.056	5	7.2	7.0	Aerated	Aerated	0	20
0.075	5	7.2	7.0			0	40
0.10	5	7.2	7.1			40	40
0.13	5	7.3	7.2			60	80
0.18	5	7.3	7.4			100	100

TABLE 40

THE EFFECTS OF TRICHLOROACETIC ACID ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
56	5	6.0	5.7	Aerated	Aerated	20	40
75	5	5.4	5.2			20	40
135	5	5.2	4.9			60	80
180	5	4.6	4.3			100	100
240	5	4.0				80	100

TABLE 41

THE EFFECTS OF 1,2,4-TRICHLOROBENZENE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
1.0	5	7.2	6.9	Aerated	Aerated	0	20
1.35	5	7.2	6.9			0	40
1.80	5	7.3	6.8			40	80
2.40	5	7.2	6.8			80	100
3.20	5	7.2	6.6			80	100

TABLE 42

THE EFFECTS OF COPPER 2-METHYL-4-CHLOROPHENOXYACETATE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 59° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
32	5	8.7	8.5	Aerated	Aerated	0	40
42	5	8.8	8.5			0	80
56	5	8.8	8.7			40	100
75	5	9.0	8.6			80	100
100	5	9.3	9.0			100	100

TABLE 43

THE EFFECTS OF COPPER SULFATE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 59° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
1.0	5	7.2	7.1	Aerated	Aerated	0	20
1.35	5	7.2	7.0			40	60
1.80	5	7.2	7.0			60	80
2.40	5	7.2	7.1			100	100
3.20	5	7.2	7.0			100	100

TABLE 44

THE EFFECTS OF COPPER DICHLOROACETATE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
10.0	5	8.6	8.3	Aerated	Aerated	0	0
11.5	5	8.6	8.4			0	40
13.5	5	8.8	8.4			20	40
15.5	5	9.1	8.7			60	80
18.0	5	9.3	9.0			100	100

TABLE 45

THE EFFECTS OF COPPER 2,4-DICHLOROPHENOXYACETATE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 64° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
10	5	7.6	7.4	Aerated	Aerated	20	40
20	5	7.6	7.5			40	60
30	5	7.7	7.5			60	100
40	5	7.7	7.5			60	100
50	5	7.7	7.4			100	100

TABLE 46

THE EFFECTS OF "AROMATIC #80" ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
3.2	5	8.2	8.0	9.6		0	0
4.2	5	8.2	8.0	9.6		20	60
5.6	5	8.2	7.9	9.5		80	100
7.5	5	8.2	7.9	9.6		80	100
10.0	5	8.3	7.9	9.4		100	100

TABLE 47

THE EFFECTS OF COPPER CHLORIDE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
0.20	5	7.8	7.7	Aerated	Aerated	0	0
0.36	5	7.8	7.7			0	0
0.64	5	7.8	7.6			40	20
1.12	5	7.8	7.7			100	20
1.50	5	7.8	7.6			100	60

TABLE 48

THE EFFECTS OF ISOPROPYL-N-PHENYLCARBAMATE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
1.0	5	7.2	7.1	9.3	9.3	0	0
1.8	5	7.3	7.2	9.3	9.1	0	40
3.2	5	7.2	7.1	9.2	9.0	40	60
5.6	5	7.2	7.0	9.2	9.1	80	100
10.0	5	7.4	7.2	9.2	9.0	100	100

TABLE 49

THE EFFECTS OF ISOPROPYL-N-PHENYLCARBAMATE (50% WETTABLE) ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
49	5	7.8	7.7	9.4	9.2	0	20
56	5	7.8	7.6	9.4	9.3	0	40
65	5	7.9	7.5	9.4	9.3	20	80
75	5	8.0	7.5	9.2	9.0	60	100
87	5	8.0	7.4	9.3	9.1	100	100

TABLE 50

THE EFFECTS OF MONOCHLOROBENZENE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
0.42	5	7.4	7.3	9.0	6.2	0	0
0.56	5	7.4	7.2	9.0	6.0	0	60
0.75	5	7.4	7.2	9.3	5.8	20	80
1.35	5	7.4	7.2	8.7	5.4	60	80
1.80	5	7.4	7.2	8.6	6.0	80	100

TABLE 51

THE EFFECTS OF PHENOXYACETIC ACID - 1% COPPER CHLORIDE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at temperature of 64° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
2	5	7.2	7.1	9.1	8.9	0	20
5	5	7.2	6.9	9.0	8.7	0	60
10	5	7.2	6.8	9.0	8.6	60	100
15	5	6.9	6.6	9.2	8.6	60	100
40	5	6.7	6.4	9.1	8.4	80	100

TABLE 52

THE EFFECTS OF PINENE ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 61° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
8.7	5	7.9		9.3		0	20
10.0	5	8.0		9.4		40	40
11.5	5	8.0		9.0		40	60
13.5	5	8.2		9.3		80	100
15.5	5	8.3		9.2		100	100

TABLE 53

THE EFFECTS OF "SOCAL #3" ON WHITE CRAPPIE.
(Tests conducted in 15 liters of solution at a temperature of 63° F.)

Concentration in p.p.m.	Number of Test Fish	pH		Dissolved Oxygen in p.p.m.		Mortality in Hours	
		Start	End	Start	End	24	48
3.2	5	7.8	7.7	9.4	8.0	0	0
4.2	5	7.8	7.5	9.3	6.2	40	40
5.6	5	7.7	7.4	9.4	5.8	40	80
7.5	5	7.8	7.3	9.1	6.2	60	80
13.5	5	7.8	7.3	9.2	6.4	60	100

STRAIGHT-LINE GRAPHICAL INTERPOLATION

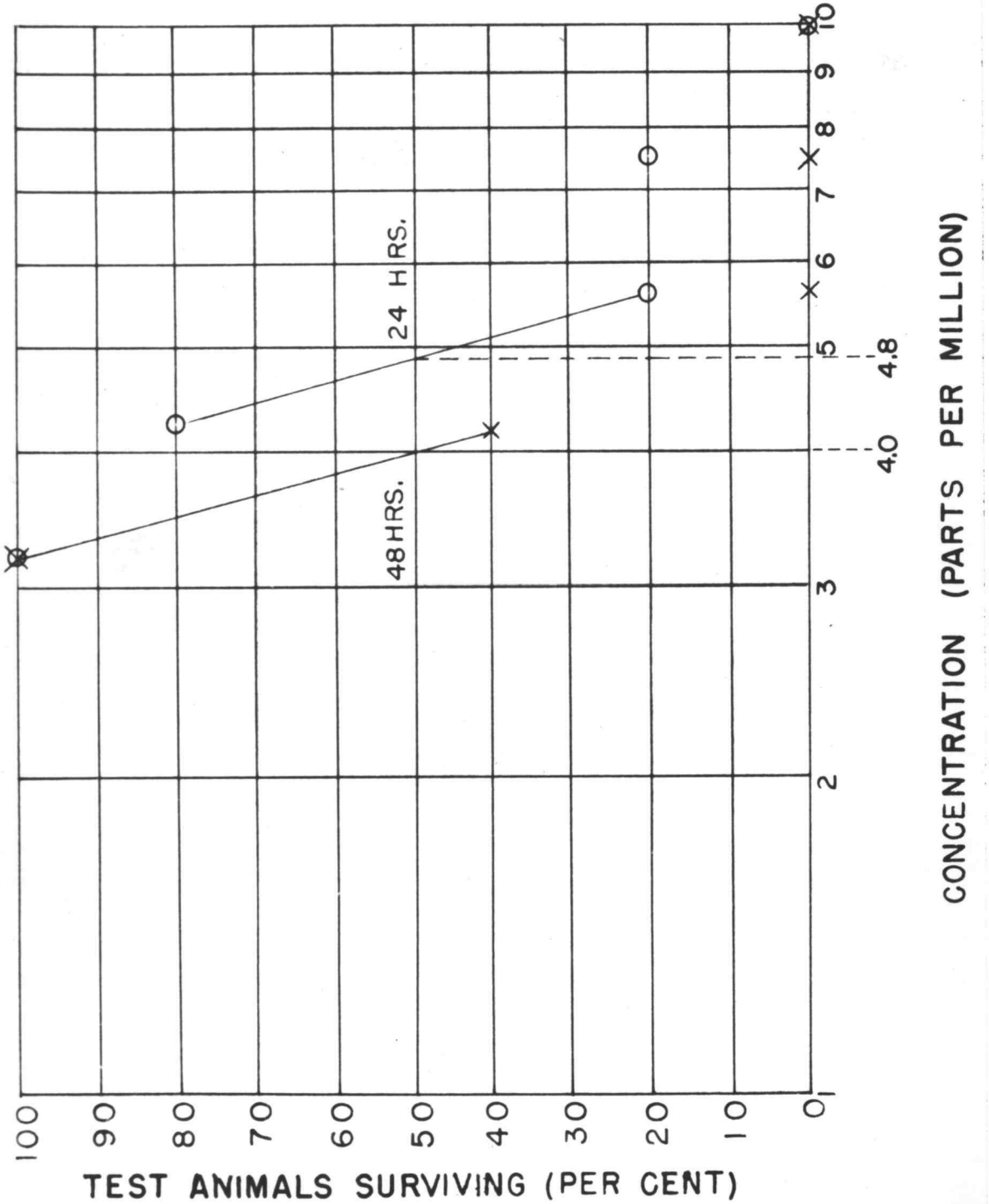
Unlike the experiments which were conducted on the mixed groups of fish in which a rough minimum lethal dosage was ascertained at the end of a 24-hour period, the evaluation of toxicity for the experiments which use only the white crappies as an exclusive test animal is expressed as a median tolerance limit (T.L.M.). (2, P. 1396) This is an index of relative toxicity at which just 50 per cent of the test animals are able to survive, for a limited period of exposure. The exposure period for these tests was 24 and 48 hours.

From figure 9 to figure 25 are a series of semilogarithmic charts on which experimental data has been plotted. The test concentrations in p.p.m. were laid off on the logarithmic scale and survival percentages on the arithmetic scale. The symbol "O" denotes the percentage of survival relative to the given concentration at the end of a 24-hour period, and the symbol "X" for a 48-hour period. To determine the median tolerance limit for one of these periods a straight line was drawn connecting two points which represent survival percentages at two successive concentrations of the test series which were lethal to more than half and to less than half of the test animals. The point at which the line intercepts the 50 per cent survival axes represents the median tolerance limit

concentration. In using a straight-line graphical interpolation of this kind it is always necessary to indicate at least one higher concentration and one lower concentration.

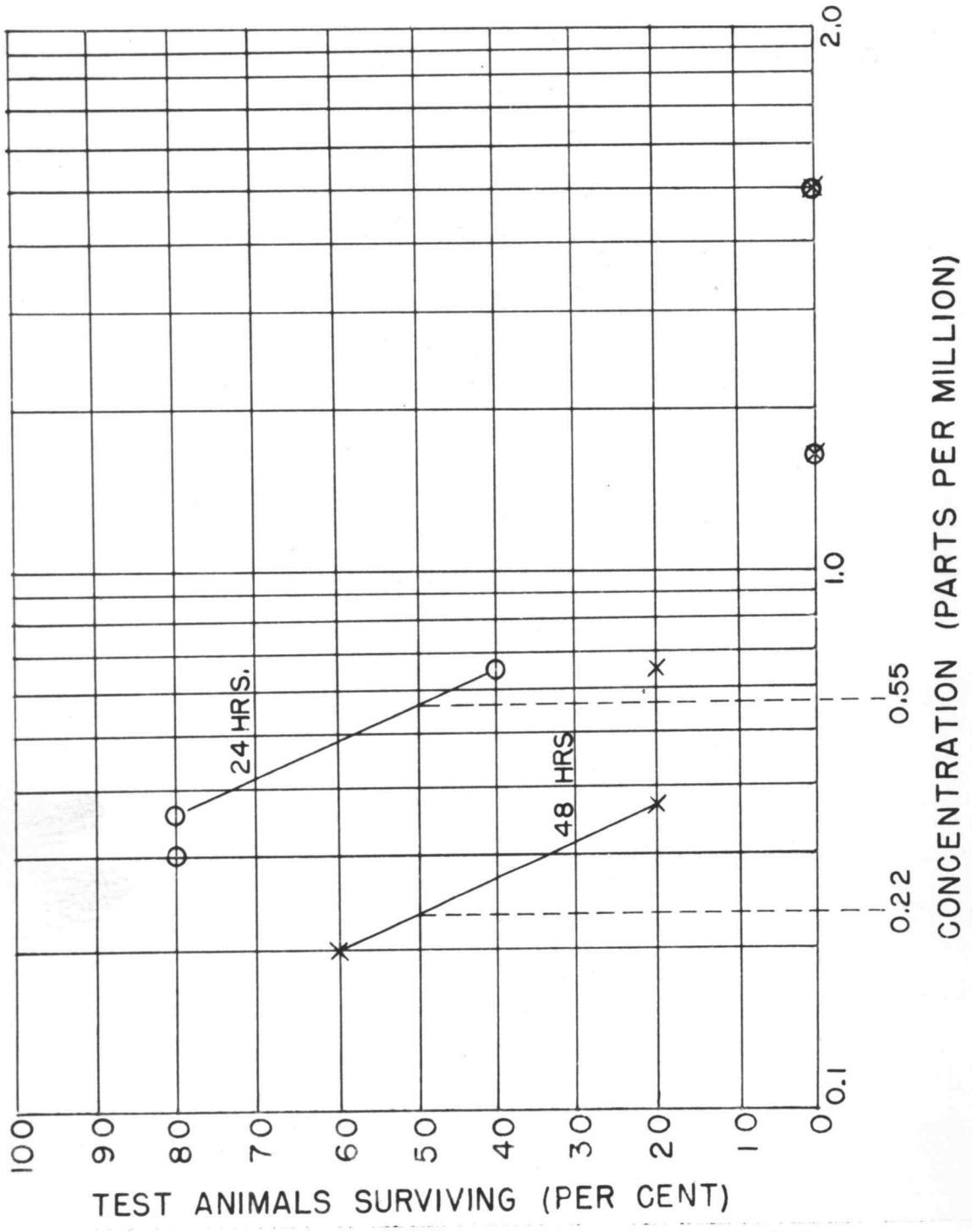
FIGURE 9

AROMATIC NO. 80



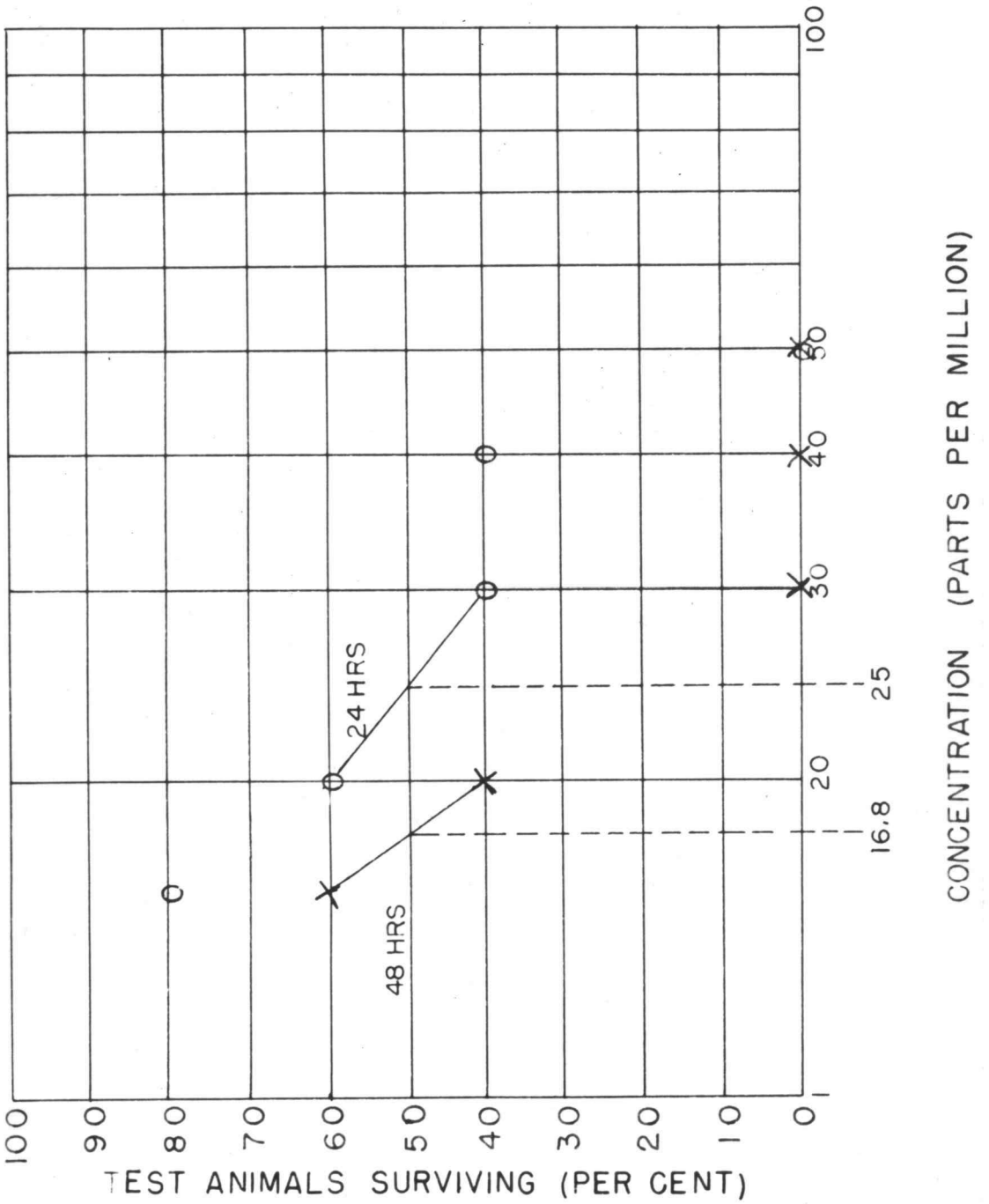
Median tolerance limits of white crappie exposed to "Aromatic #80". TL_m for 24 hours = 4.8 p.p.m.; TL_m for 48 hours = 4.0 p.p.m.

FIGURE 10
COPPER CHLORIDE



Median tolerance limits of white crappie exposed to copper chloride. TL_m for 24 hours = 0.55 p.p.m.; TL_m for 48 hours = 0.22 p.p.m.

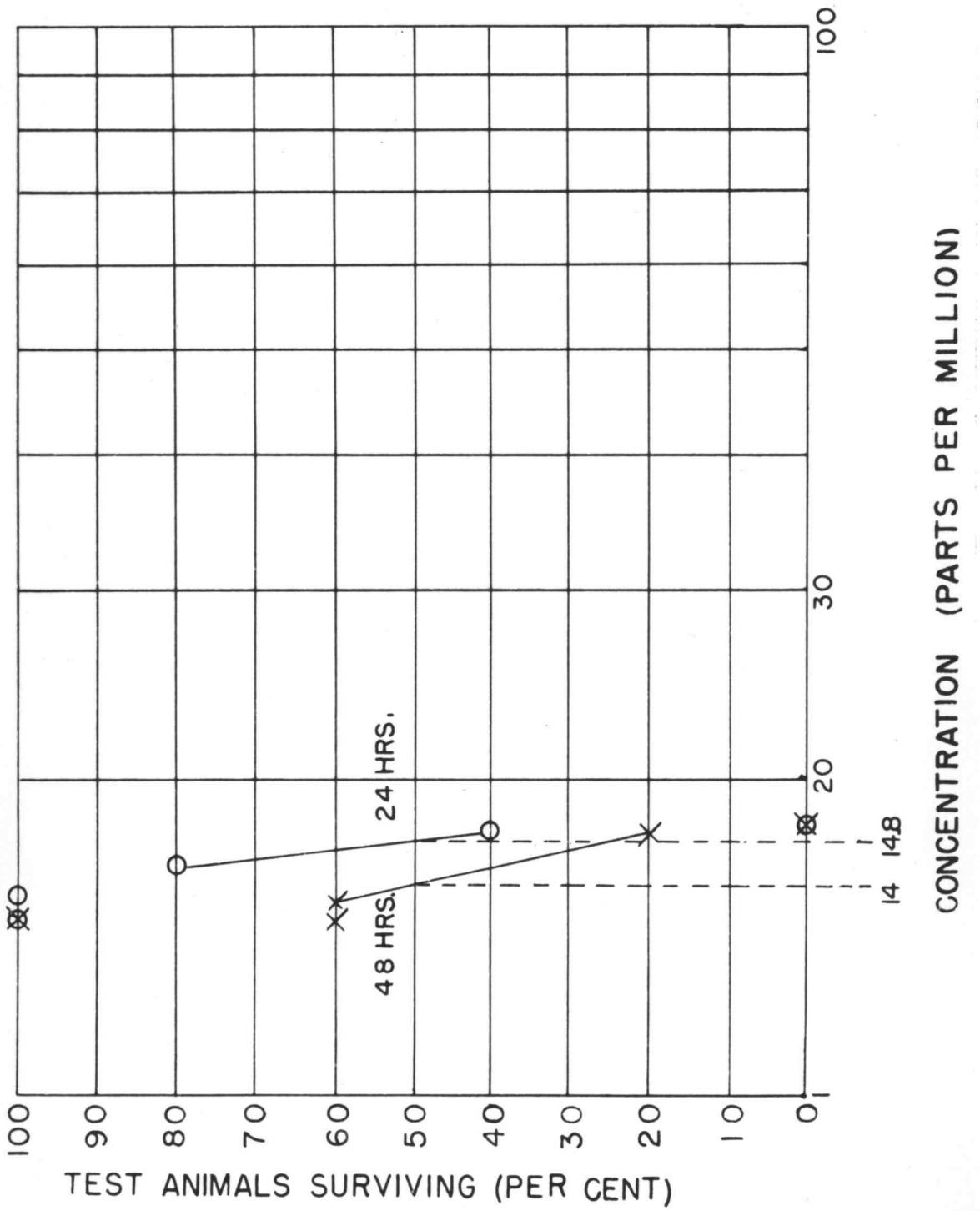
FIGURE 11
COPPER 2, 4-D



Median tolerance limits of white crappie exposed to copper 2,4-D. TL_m for 24 hours = 25 p.p.m.; TL_m for 48 hours = 16.8 p.p.m.

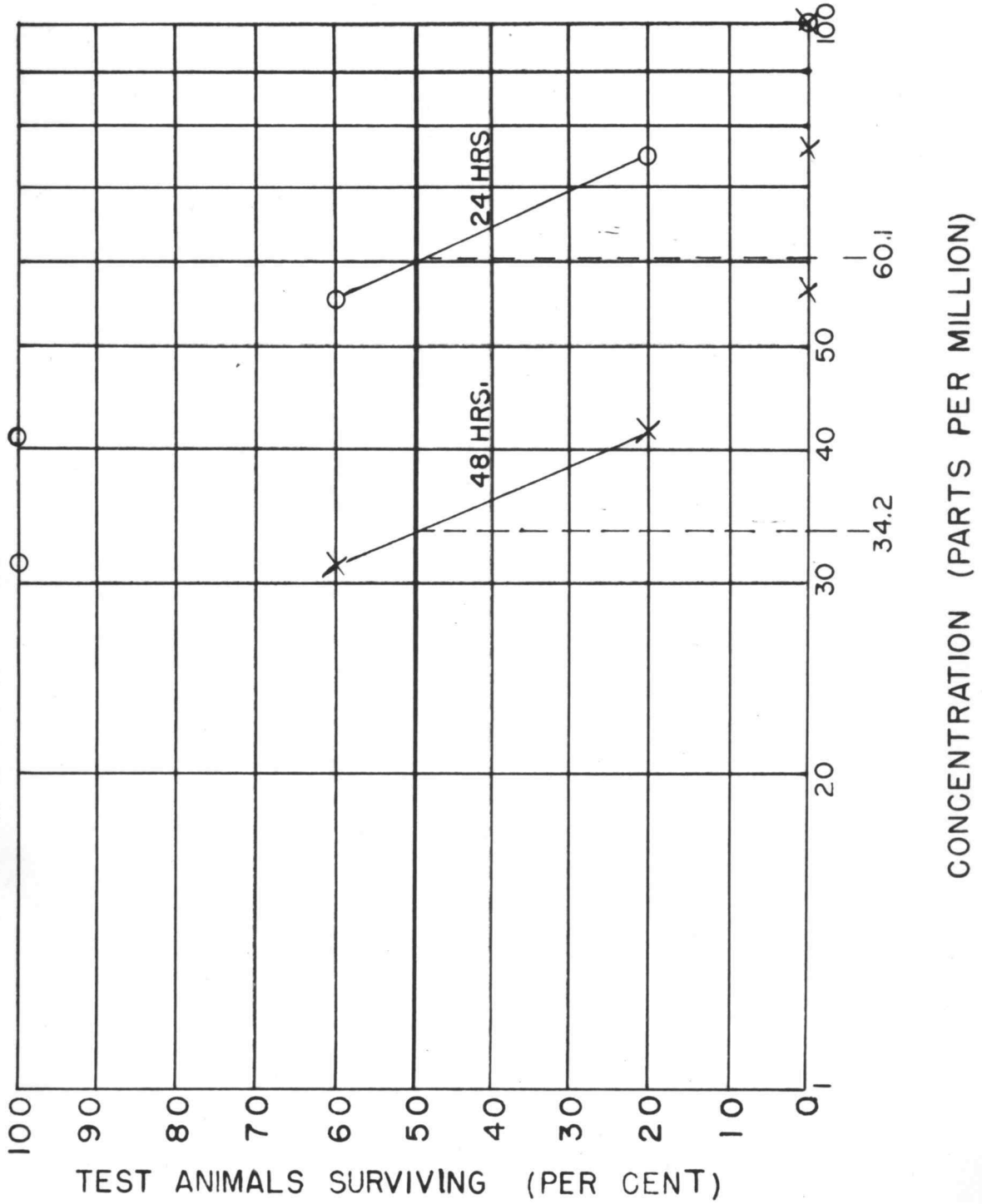
FIGURE 12

COPPER DICHLOROACETATE



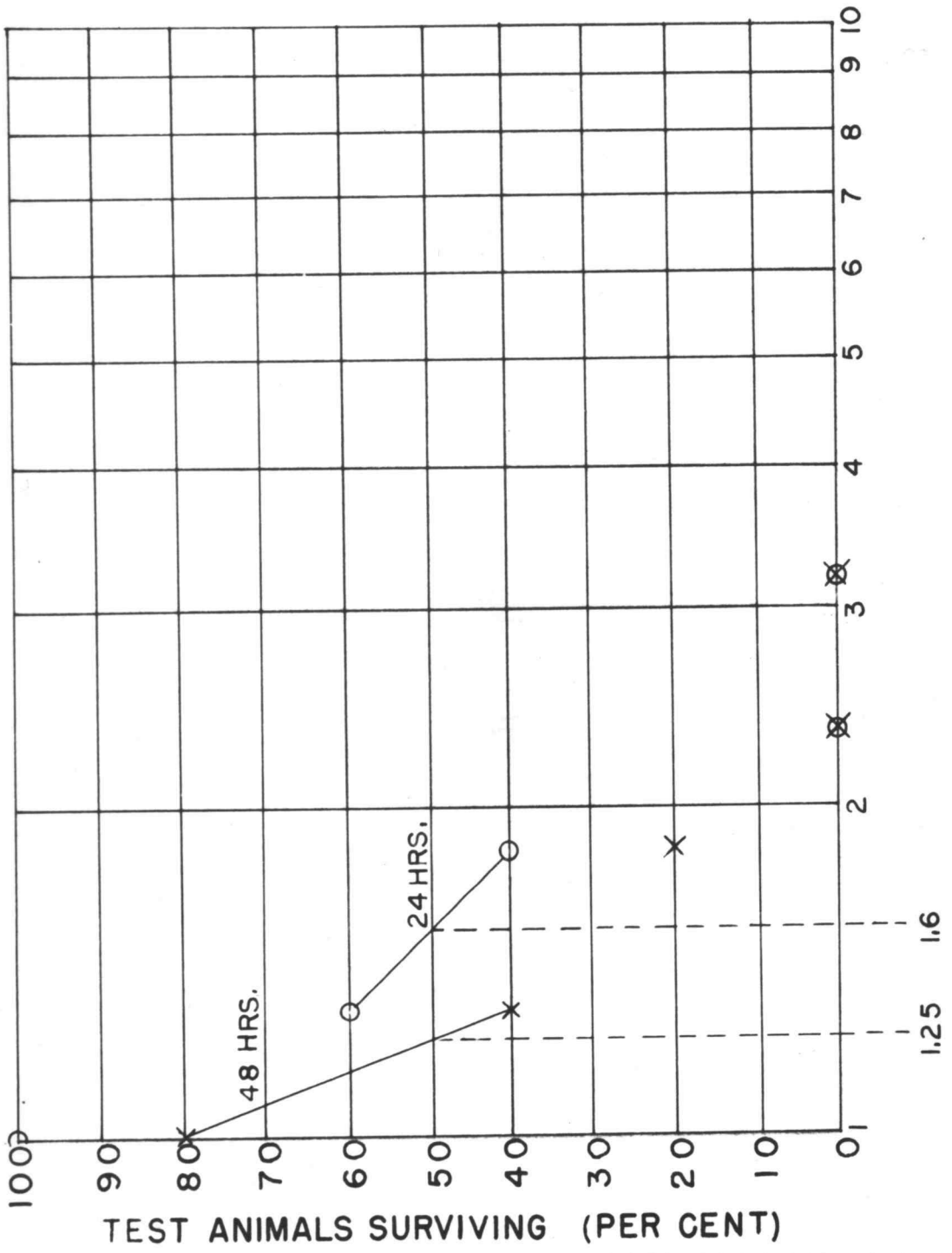
Median tolerance limits of white crappie exposed to copper dichloroacetate. TL_m for 24 hours = 14.8 p.p.m.; TL_m for 48 hours = 14. p.p.m.

FIGURE 13
COPPER SALT OF MCPA



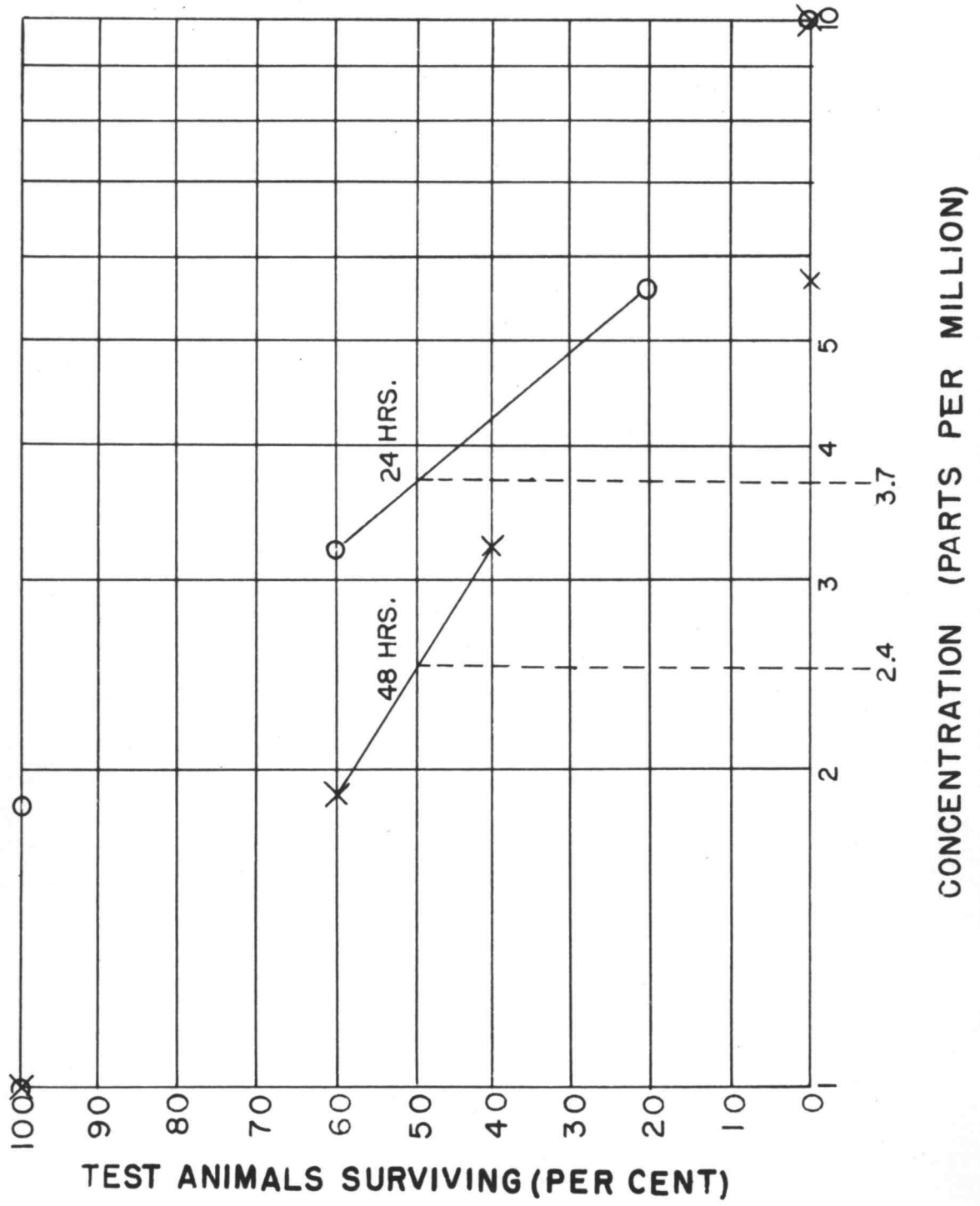
Median tolerance limits of white crappie exposed to copper MCPA. TL_m for 24 hours = 60.1 p.p.m.; TL_m for 48 hours = 34.2 p.p.m.

FIGURE 14
COPPER SULFATE



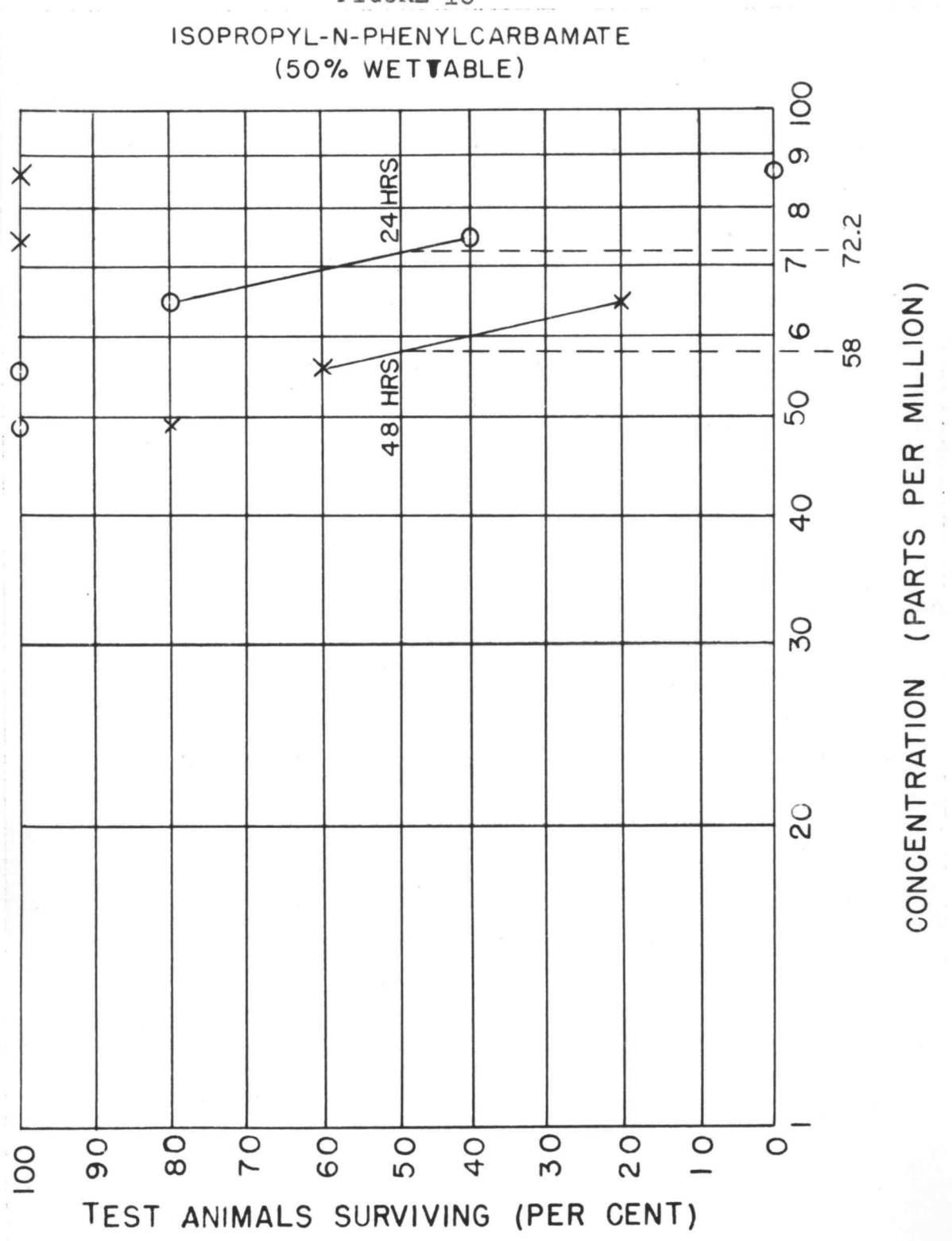
Median tolerance limits of white crappie exposed to copper sulfate. TL_m for 24 hours = 1.6 p.p.m.; TL_m for 48 hours = 1.25 p.p.m.

FIGURE 15
ISOPROPYL-N-PHENYLCARBAMATE



Median tolerance limits of white crappie exposed to isopropyl-N-phenylcarbamate. TL_m for 24 hours = 3.7 p.p.m.; TL_m for 48 hours = 2.4 p.p.m.

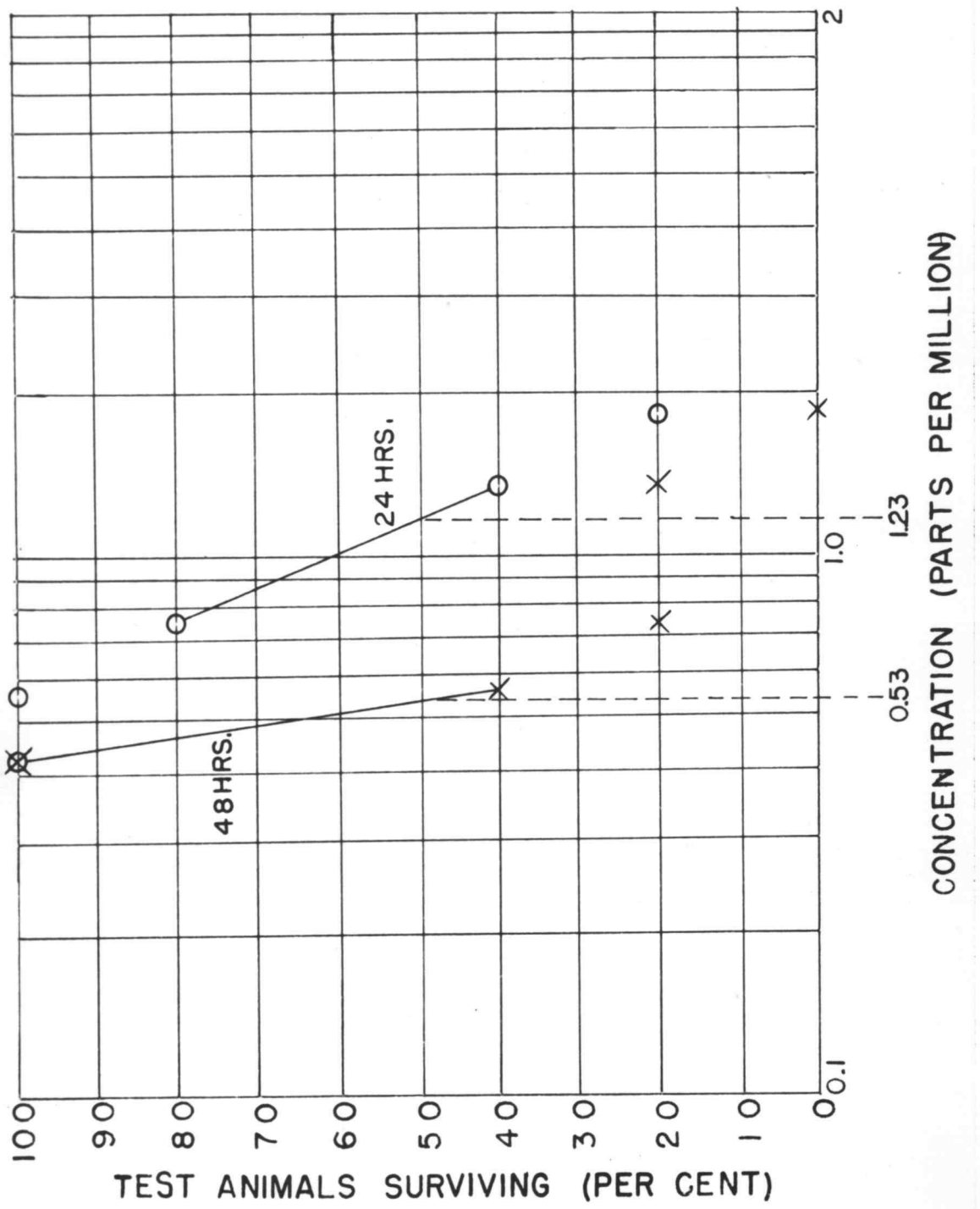
FIGURE 16



Median tolerance limits of white crappie exposed to isopropyl-N-phenylcarbamate (50% wettable). TL_m for 24 hours = 72.2 p.p.m.; TL_m for 48 hours = 58 p.p.m.

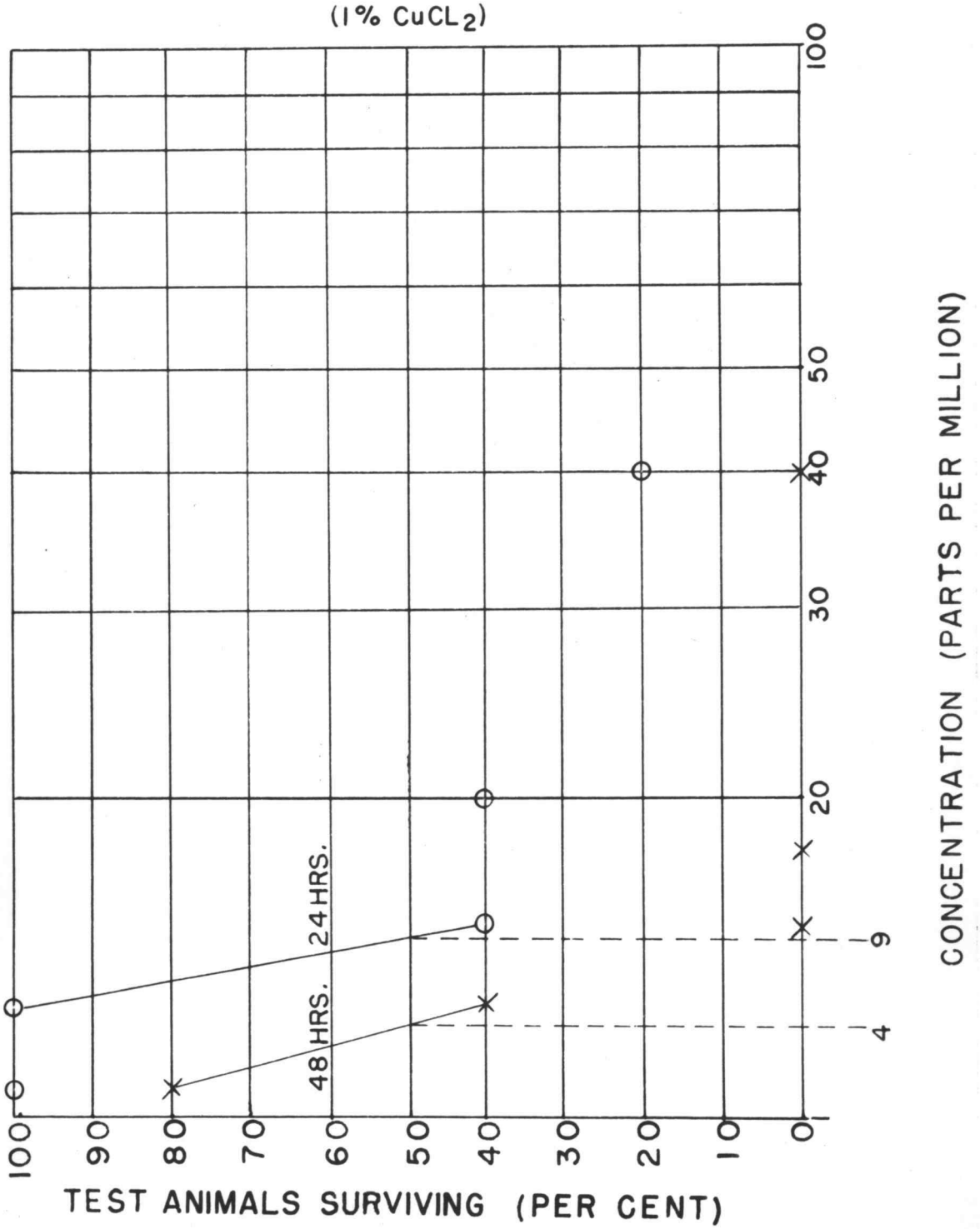
FIGURE 17

MONOCHLOROBENZENE



Median tolerance limits of white crappie exposed to monochlorobenzene. TL_m for 24 hours = 1.23 p.p.m.; TL_m for 48 hours = 0.53 p.p.m.

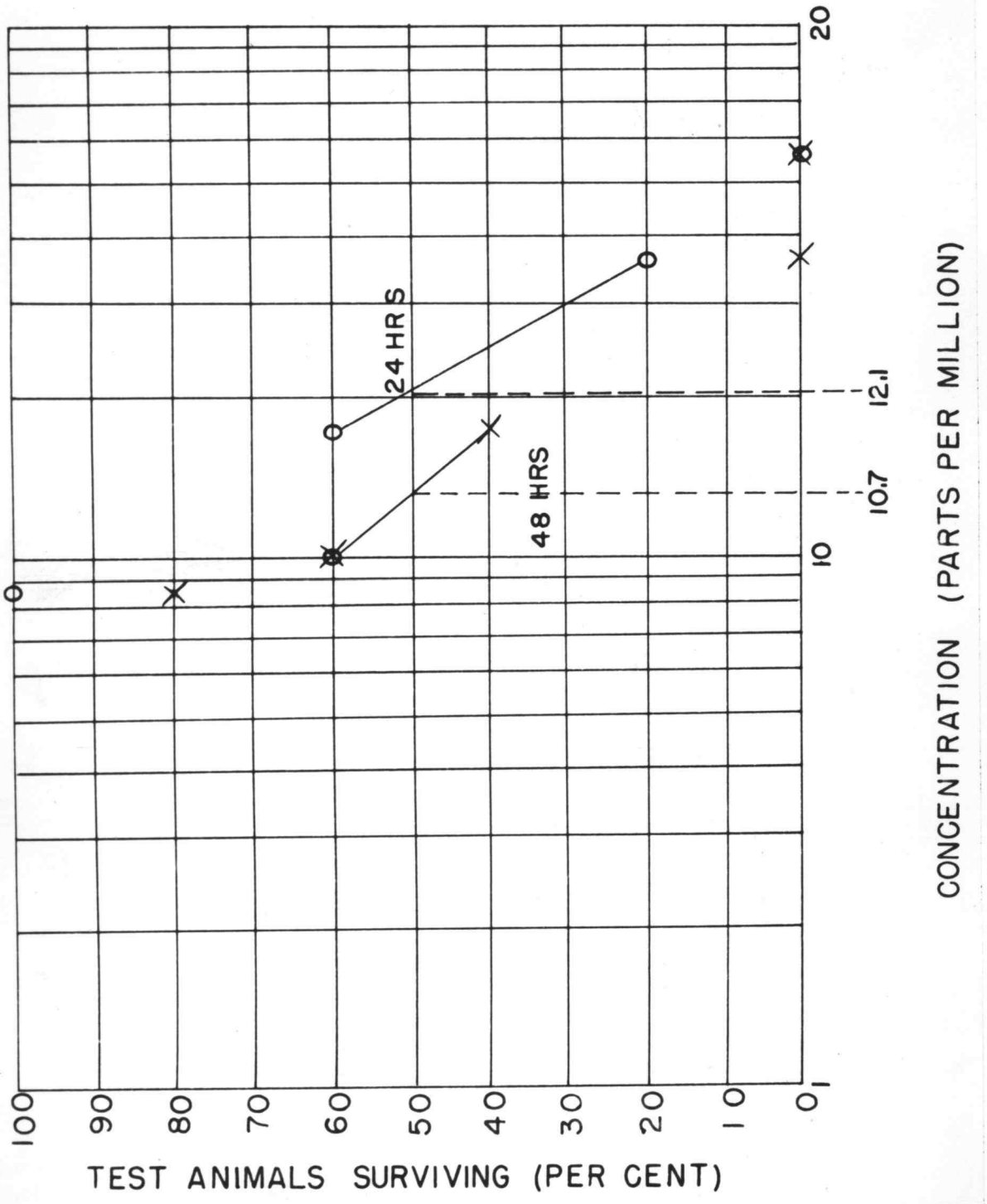
FIGURE 18



Median tolerance limits of white crappie exposed to phenoxyacetic acid (1% $CuCl_2$). TL_m for 24 hours = 9 p.p.m.; TL_m for 48 hours = 4 p.p.m.

FIGURE 19

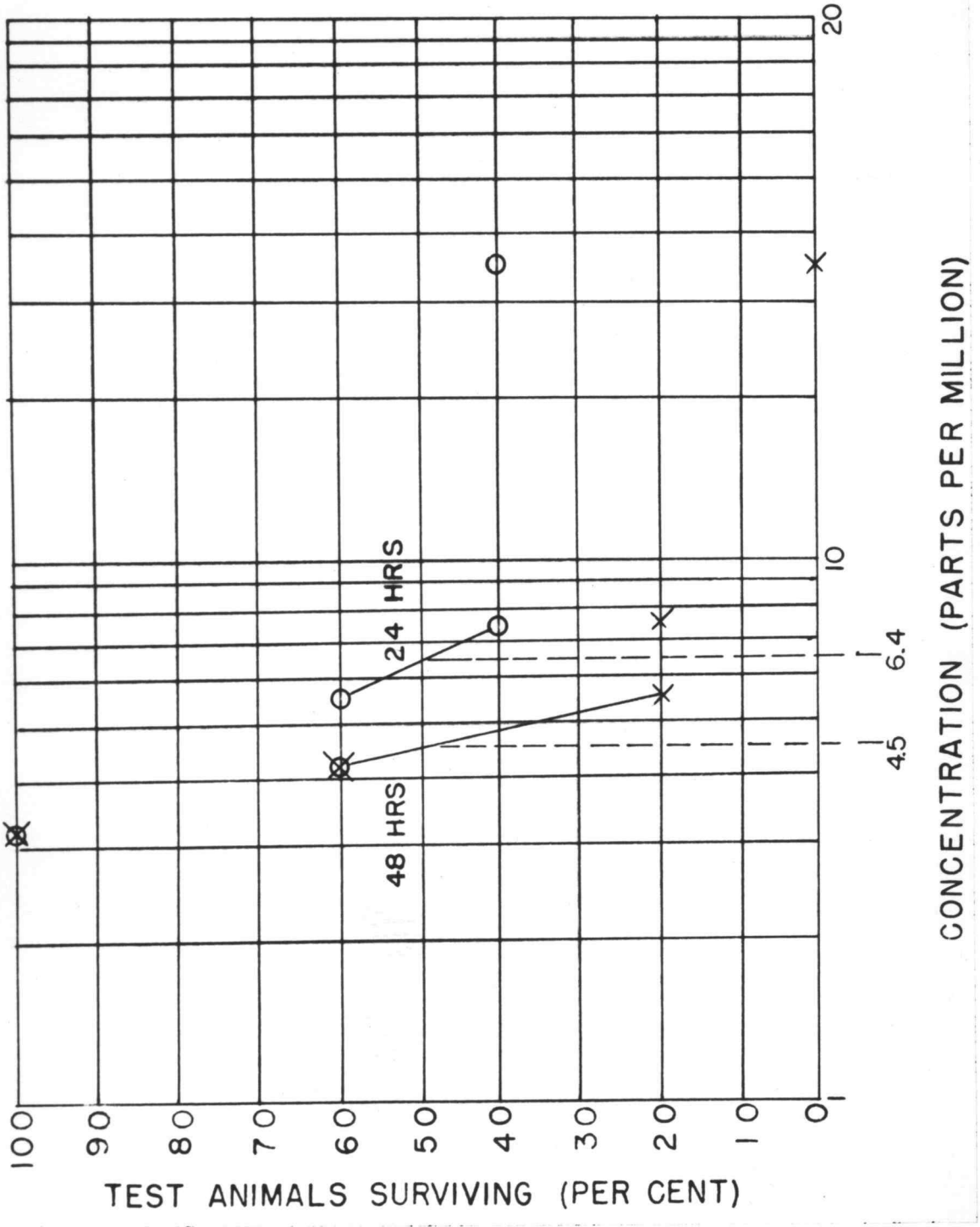
PINENE



Median tolerance limits of white crappie exposed to pinene. TL_m for 24 hours = 12.1 p.p.m.; TL_m for 48 hours = 10.7 p.p.m.

FIGURE 20

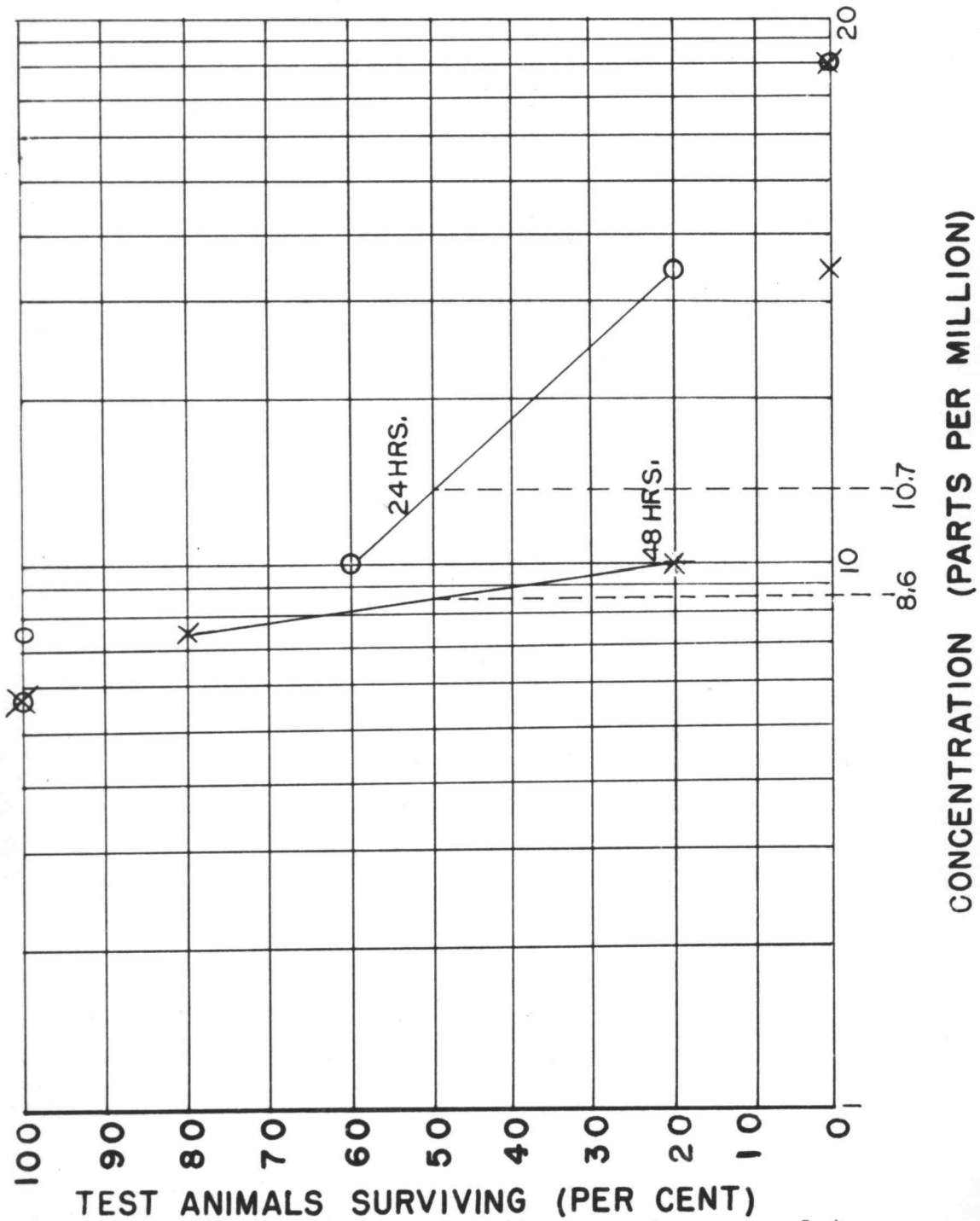
SOCAL NO. 3



Median tolerance limits of white crappie exposed to "Socal #3". TL_m for 24 hours = 6.4 p.p.m.; TL_m for 48 hours = 4.5 p.p.m.

FIGURE 21

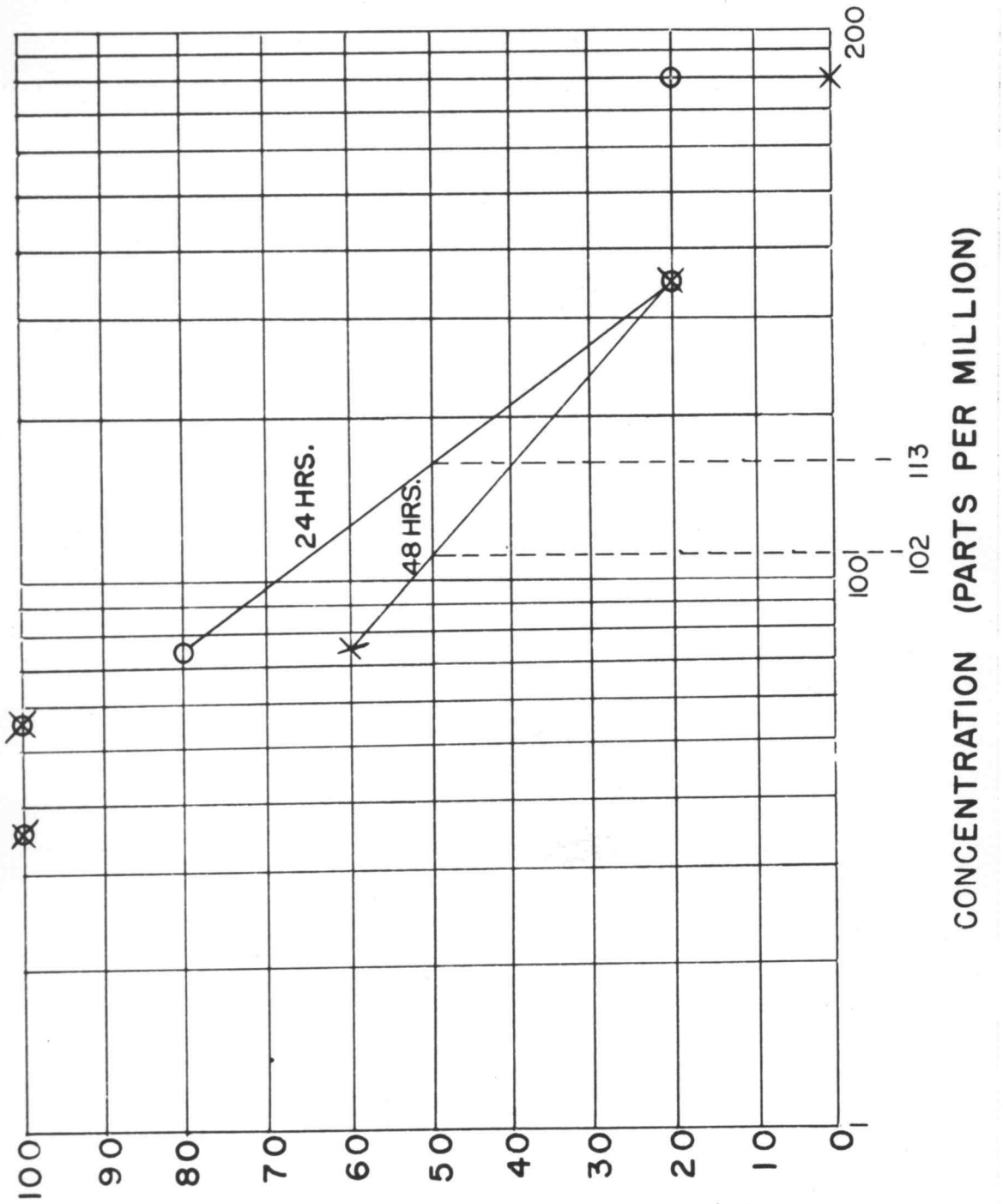
SODIUM 2,4-DINITROPHENATE



Median tolerance limits of white crappie exposed to sodium 2,4-dinitrophenate. TL_m for 24 hours = 10.7 p.p.m.; TL_m for 48 hours = 8.6 p.p.m.

FIGURE 22

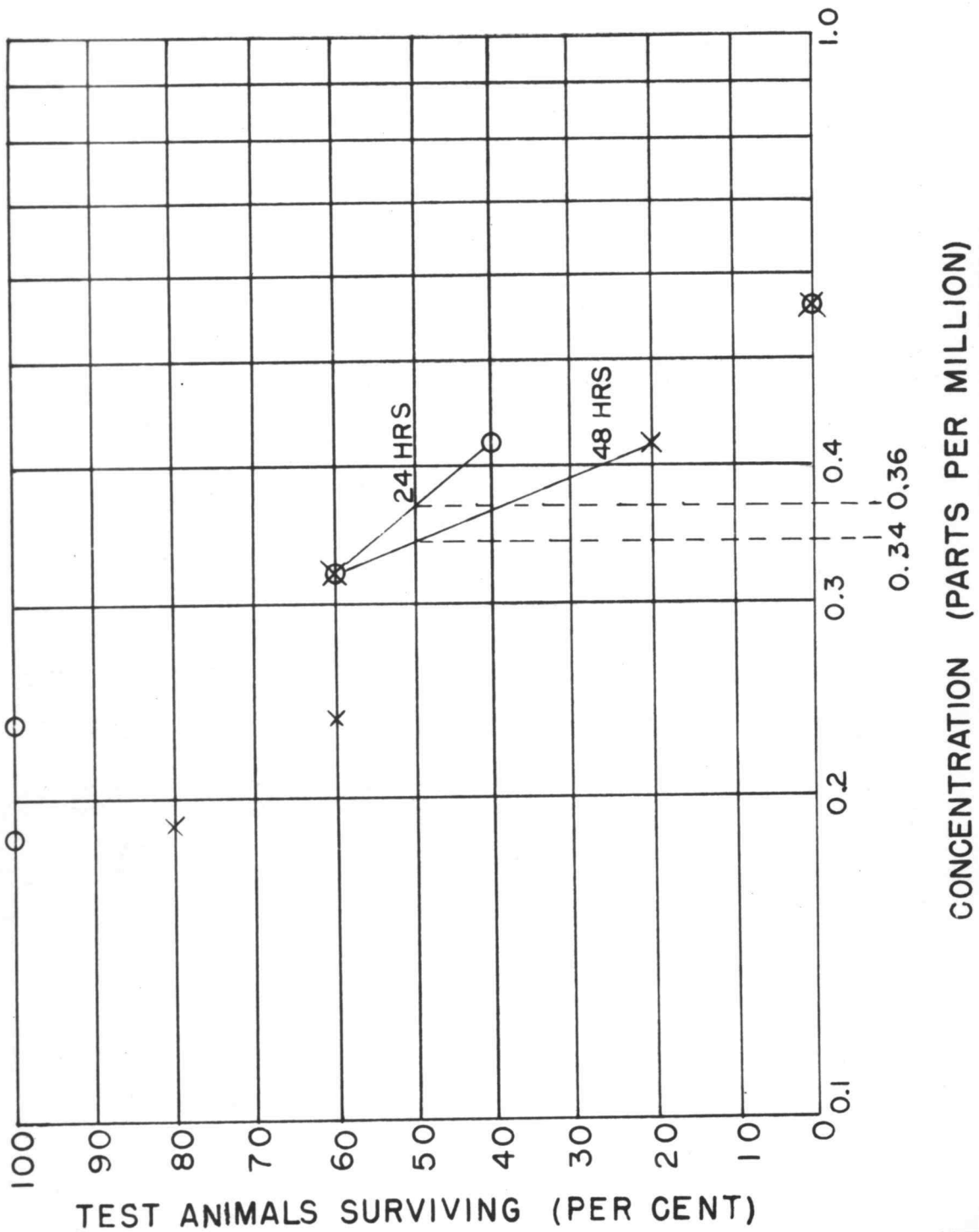
SODIUM MCPA



Median tolerance limits of white crappie exposed to sodium MCPA. TL_m for 24 hours = 113 p.p.m.; TL_m for 48 hours = 102 p.p.m.

FIGURE 23

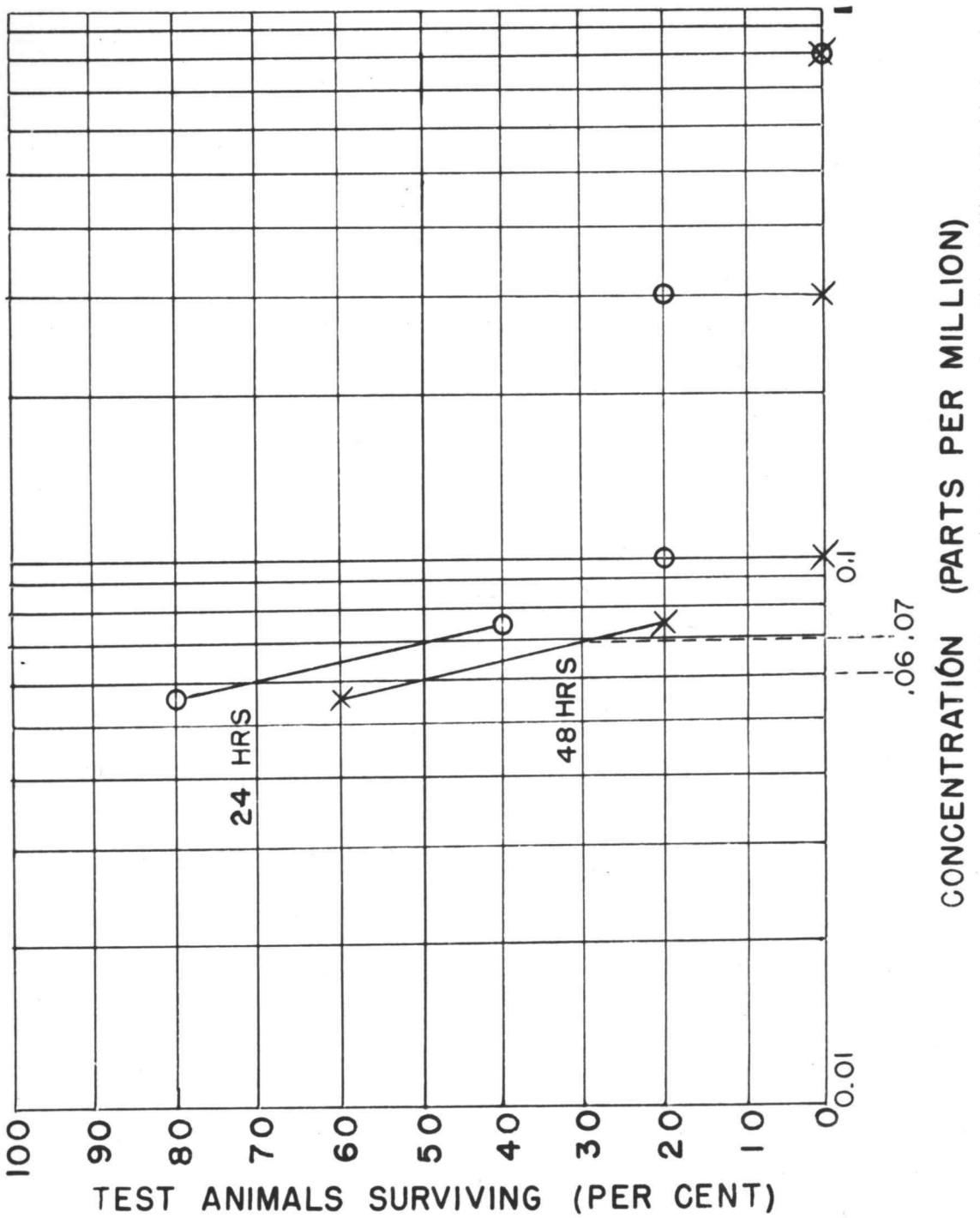
SODIUM PENTACHLOROACETATE



Median tolerance limits of white crappie exposed to sodium pentachloroacetate. TL_m for 24 hours = 0.36 p.p.m.; TL_m for 48 hours = 0.34 p.p.m.

FIGURE 24

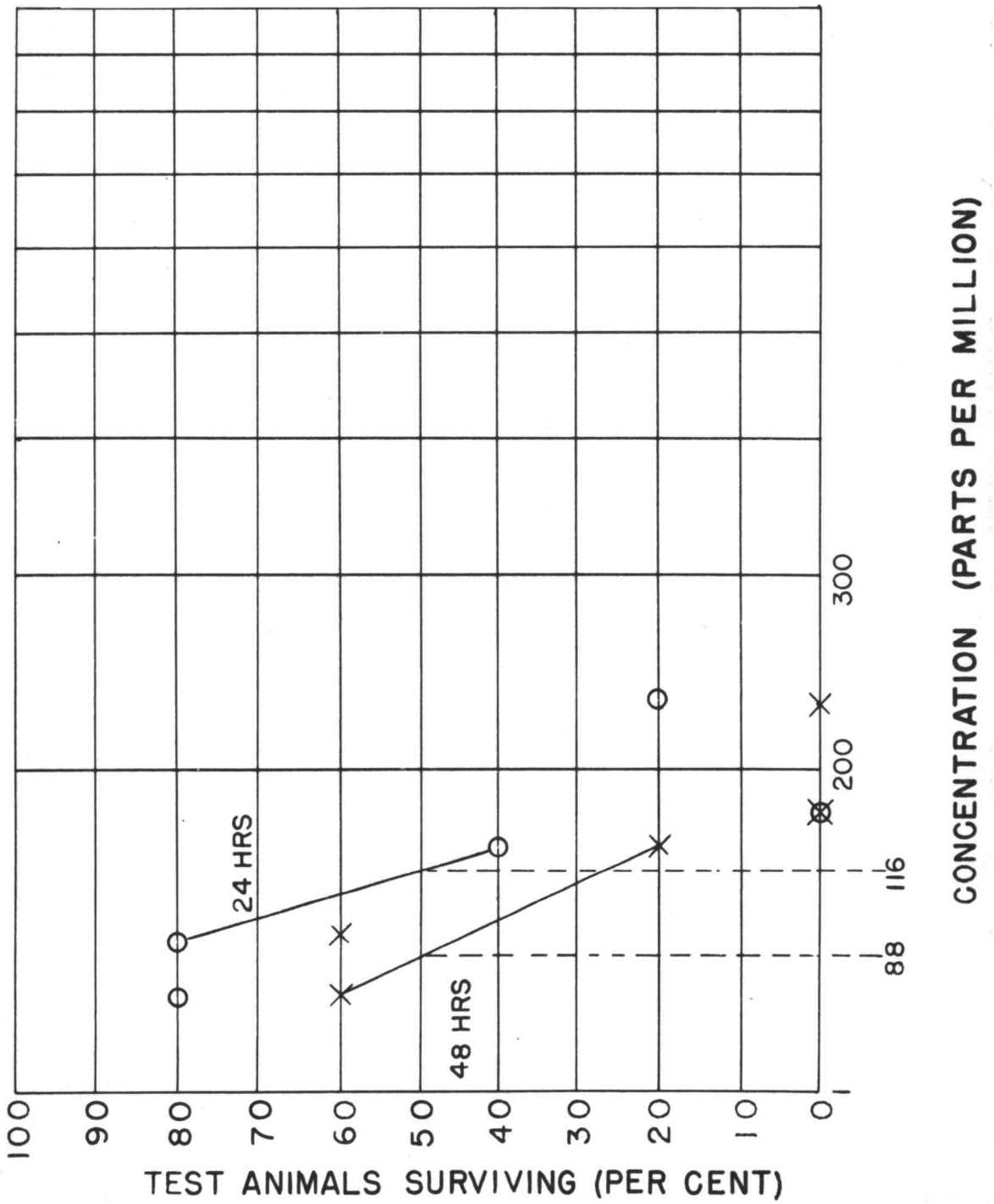
SODIUM PENTACHLOROPHENATE



Median tolerance limits of white crappie exposed to sodium pentachlorophenate. TL_m for 24 hours = 0.07 p.p.m.; TL_m for 48 hours = 0.06 p.p.m.

FIGURE 25

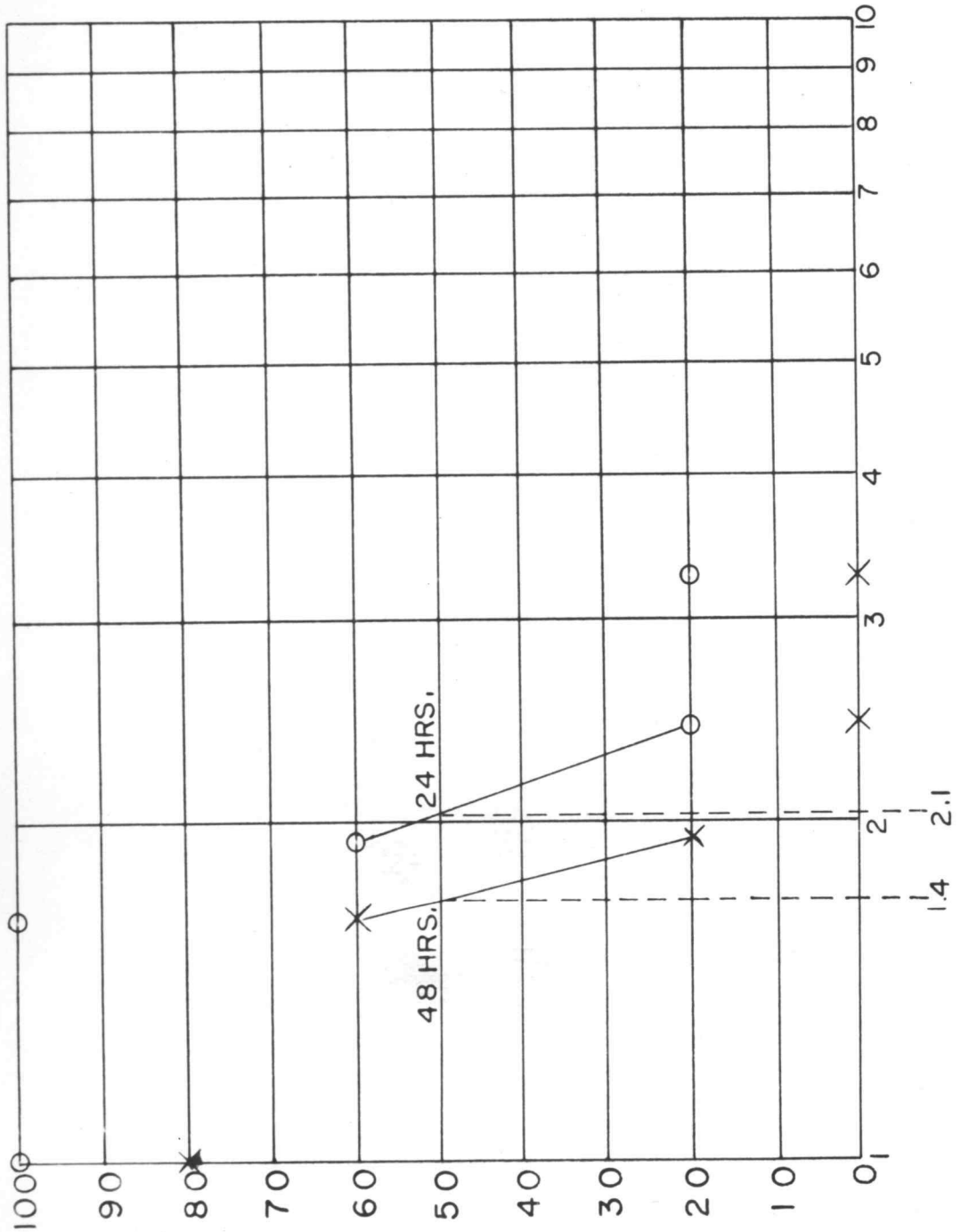
TRICHLOROACETIC ACID



Median tolerance limits of white crappie exposed to trichloroacetic acid. TL_m for 24 hours = 116 p.p.m.; TL_m for 48 hours = 88 p.p.m.

FIGURE 26

1,2,4-TRICHLOROBENZENE



Median tolerance limits of white crappie exposed to 1,2,4-trichlorobenzene. TL_m for 24 hours = 2.1 p.p.m.; TL_m for 48 hours = 1.4 p.p.m.

CONCLUSION

By and large most of the copper salts of organic acids used in these experiments gave the most promising results as an effective herbicide for Anacharis densa. These salts, for the most part, would kill the water weed at a concentration which was not lethal to the fish.

The minimum lethal dosage of copper dichloroacetate on white crappie, bluegill and largemouth bass was roughly 30 p.p.m., table 6. It was noted that some of the copper precipitated out in the mucosa that was sloughed off by the test animals.

Copper monochloroacetate did not produce an M.L.D. on the fish at 100 p.p.m., but made just as an effective kill on the water weed as copper dichloroacetate, table 9. It would seem that this compound would be the more desirable of the two as a herbicide.

Copper MCPA made a 100% kill on white crappie between 42-56 p.p.m., figure 28. Since this compound would kill the water weed at a concentration of 4 p.p.m. when dissolved in "Socal #3", it should then warrant further investigation. Furthermore, when "Socal #3" is used in a killing concentration by itself, it produces a high mortality on fish. When copper MCPA was used to fortify "Socal #3", however, actual trial on Siltcoos Lake showed no evidence of fish being killed.

The copper salt of phenylacetic acid is easy to formulate and it solubilizes readily in concentrated ammonium hydroxide. Toxicity experiments showed that its M.L.D. is approximately 75 p.p.m., table 32. Since it is capable of killing the water weed in test jars at 5 p.p.m., it should prove possibly one of the more economic herbicides in the event that material of this sort is ever used for this purpose.

The copper salt of 2,4-D acted similarly to copper MCPA on both the white crappie and the water weed. It produced an M.L.D. of 50 p.p.m. at the end of 24 hours, table 48. This compound goes into solution readily in concentrated ammonium hydroxide, but failed to show any appreciable toxicity on the water weed when used on field studies. Unlike Cu MCPA, it would not dissolve in "Socal #3", and hence could not be used for fortifying aromatic solvents.

There were, in addition to the metallic compounds previously mentioned, several other copper salts used in these experiments. Most of these were formulated from organic acids, phenolic compounds and alcohols which were on hand at the laboratory on the South Farm. For the most part these salts were just as toxic to the water weed as the commercial growth regulators, but they were considerably more toxic to the fish because of the amount of

concentrated ammonium hydroxide needed to take them into solution. It would therefore seem that unless another solvent could be found for this material, its general use as a herbicide could not be encouraged.

The sulphonated compounds were, in particular, difficult to get into solution with any solvent, and are therefore of questionable value.

The heterocyclic groups were toxic at very low concentrations. Camphene killed white crappie, bluegill, and largemouth bass at 2-5 p.p.m., table 18. Pinene was lethal to the same fish at 5-10 p.p.m., table 20, and 12.1 p.p.m. killed white crappie when it was used as an exclusive test animal, figure 19.

The sodium salt of 2,4-D was not particularly toxic to white crappie. This fish suffered a 40% mortality at a concentration of 1350 p.p.m., and an M.L.D. at 1500 p.p.m., figure 11. Sodium MCPA produced an M.L.D. at 300 p.p.m., figure 13.

Emulsifying reagents which were needed for the less water soluble compounds, varied considerably in their ability to toxify fish. "Tween-20" produced no ill effects on white crappie at a concentration of 300 p.p.m. Since this was the highest eoncentration used on this fish there is no way of knowing just where the M.L.D. lies. Triethanolamine showed no visible effects on white

crappie, bluegill or largemouth bass at 500 p.p.m. "Trex-40" was lethal to these same fish at a concentration of 5-25 p.p.m. "Tenlo-400", which was probably the most toxic of all the emulsifiers, killed bluegill and white crappie at 4 p.p.m.

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