

EVALUATION OF ANHYDROUS AMMONIA FOR FISHERY MANAGEMENT USES¹

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ABSTRACT

Anhydrous ammonia was applied to ponds at average rates of 18-40 ppm to evaluate its use as a fish toxicant and aquatic herbicide. Ponds contained toxic levels of ammonia for 3-4 weeks after applications, depending on concentration, water temperature and other factors. A regression model was developed to estimate specific dissipation times. Application of 15 ppm ammonia effected a high percent mortality to all fish species observed. Complete fish kills occurred at approximately 30 ppm ammonia. Any concentration above 15 ppm temporarily controlled most vegetation, regardless of season. Benthic organisms were reduced but not eliminated by any treatment. Anhydrous ammonia was found to be relatively inexpensive compared to other fish toxicants and aquatic herbicides.

This study evaluates anhydrous ammonia as a fish toxicant and aquatic herbicide. Its effects on the benthic community and water quality were also examined.

In pond management it is often necessary to remove or reduce undesirable fish and aquatic plant populations; however, many toxins and herbicides presently used leave a persistent nonbiodegradable residue. Nitrogen and its compounds are present throughout the environment, and in aquatic systems they are most prevalent as by-products of metabolism. Under certain conditions, in sufficient concentrations, some nitrogen compounds can be harmful to aquatic life.

Effects of nitrogen compounds on aquatic organisms have been studied for years. Ramachandron (1960) reported that 18 ppm ammonia killed several species of emergent vegetation, and such submerged species as *Hydrilla verticillata* and *Najas* spp. within 5 days. Benthic organisms were also killed but reestablished rapidly. Concentrations of 20 and 30 ppm gave similar results.

Duodoroff and Katz (1950) indicated concentrations from 2-7 ppm ammonia to be lethal to fish. Clark and Adams (1913) reported 13 ppm killed minnows and suckers. Shelford (1917) found 7-8 ppm killed orange-spotted sunfish in 1 hr.

Klussman, Champ, and Lock (1969) tested several ammonia concentrations on 15 Central Texas ponds. They found ammonia useful as a fertilizer, fish toxicant, and herbicide, and certain concentrations might be selective in killing certain species of fish.

We wish to express appreciation to the U. S. Fish and Wildlife Service for support of this study under the Federal Aid in Fish and Wildlife Restoration Program. Special credit is due Dr. Wallace G. Klussmann of Texas A&M University and Mr. William P. Rutledge of the Texas Parks and Wildlife Department for their efforts in supervising segments of field work. Appreciation is expressed for the efforts of Mr. Archie Lee, Jr. and Mr. Warren Wagner for many hours of field work, and to personnel of the Data Processing Division for thorough analyses of the data. Thanks also go to various Department personnel for reviewing the manuscript.

¹ Contribution from Federal Aid Restoration Funds under Derigell-Johnson F-31-R.

MATERIALS AND METHODS

Four ponds measuring 0.74, 0.69, 1.05, and 0.96 ha were located on the Heart of the Hills Research Station in Kerr County, Texas, operated by the Texas Parks and Wildlife Department. Each was treated with concentrations meant to approximate either 10, 20, 30, or 40 ppm anhydrous ammonia. Treatments were repeated four times over 2 years allowing for 2 summer and 2 winter replications. Concentrations were assigned to ponds randomly for each treatment. Ponds were filled 2 months before treatment in an effort to allow ecosystem stabilization.

Ammonia was applied with a portable pressure tank (similar to those used in agricultural fertilizing) at a rate of 1 g NH₃/m³ water for 1 ppm of treatment desired. The tank was equipped with a pneumatic scale for weighing the chemical. Three high pressure hoses, (1.6 cm outside diameter), tipped with perforated stainless steel probes (1.2 m in length) dispersed the chemical. These probes were held vertically in the water column, approximately equidistant apart, for maximum dispersion.

Water quality parameters were measured at least twice before treatment, and at various intervals after treatment, until ammonia reached pretreatment levels. Water samples were taken with a Kemmerer sampler at a depth of 1 m. Parameters measured and techniques used were: 1) temperature and oxygen, Delta Model 75; 2) pH, Corning Model 10; 3) alkalinity, standard acid titration; 4) total hardness, EDTA titration; 5) conductivity, Beckman SOLU Bridge RB-3; 6) turbidity, Jackson turbidity units; 7) ammonia, direct Nesslerization; and 8) nitrate-nitrite, cadmium reduction method. Those measurements not made with an electronic instrument were made with a Hach DR-EL "direct reading" portable water analysis kit.

Since the number of samples taken during each test varied, data were divided into four time periods for analyses. Time periods were defined as: Time I, before ammonia application; Time II, 1-5 days after application; Time III, 6-10 days after application; and Time IV, 11-15 days after application. Data were analyzed using a three-way factorial ANOVA with unequal cell sizes (Overall and Spiegel 1969). Each parameter was tested for differences between years, time periods, and ammonia concentrations. Multiple regression analysis was used to develop a method to predict how long ammonia, under certain conditions, remained in an ecosystem.

Benthic samples were taken once before and several times after treatment. Four Ekman dredge samples from various locations in each pond were combined, screened through a 30-mesh sieve and the organisms preserved. Each combined sample represented 0.09 m² of bottom fauna. Organisms were identified and counted.

Aquatic plants were inspected visually before and after treatments using a snorkel and mask or submersible viewbox. Species were identified and percent bottom cover by each estimated. Vegetation and benthos sampling were on identical days.

Benthos and vegetation results were divided into three time periods: Time I, before ammonia application; Time II, 1-33 days after application; and Time III, 34-60 days after application. Data were analyzed using a three-way factorial ANOVA with unequal cell sizes. The analyses tested for differences between years, time periods, and ammonia concentrations.

Effects of ammonia applications of fishes were determined by bioassays. Five live boxes were placed in each pond 24 hrs before treatment. Four boxes were located near the midpoint of each shoreline of the rectangular ponds and the fifth box near the bottom in the deepest area. Species stocked in each box always included members of catfish, sunfish, sucker, and minnow families. When possible, 10 individuals of each species were stocked in each box. Fishes were obtained from state fish hatcheries and private fish farms.

Live box observations were made at 6 hr intervals, for 72 hr or until all fish died. Seasonal replications were combined, giving one set of mortality values for each ammonia concentration applied in winter and one set for each concentration in summer. Accumulative mortalities were determined by dividing the total accumulated dead fish observed at each interval by total number of fish stocked. Mortalities were obtained for all species combined and individually. Regression lines were fitted indicating effects of ammonia on accumulative mortality. The model used was:

Accumulative Mortality (%) = $b_0 + b_1$ (Elapsed time), were:

b_0 = y intercept and

b_1 = regression coefficient (slope).

Regression lines were tested using covariance analysis to determine differences between seasons and ammonia concentrations.

RESULTS

Ammonia application rates were originally set to approximate 10, 20, 30, and 40 ppm (treatments 1, 2, 3, and 4 respectively). Variation occurred between observed and expected concentrations (Table 1). Cause of the variation was unknown, but a malfunction in the metering system possibly delivered more ammonia than desired for the lower concentrations.

Table 1. Ammonia concentrations immediately after application in all four ponds at Heart of the Hills Research Station, 1971-73.

Treatment	Expected Concentration (ppm)	Actual Concentration				Treatment Mean
		Summer 1971	Winter 1972	Summer 1972	Winter 1973	
1	10	22.0	15.7	16.6	16.9	17.8
2	20	27.1	26.3	19.9	31.8	26.3
3	30	30.4	38.3	27.5	35.3	32.9
4	40	41.7	39.3	37.1	38.9	39.3

Water Quality

All application rates significantly lowered dissolved oxygen in pond waters. Dissolved oxygen returned to pretreatment levels after 10 days. Summer pretreatment levels ranged from 4.9-9.1 ppm. After 5 days values ranged from 0.4-3.7 ppm. In all ponds but those receiving Treatment 1, levels dropped low enough to kill most fishes. Winter-dissolved oxygen levels reached their low point about 10 days after treatment. Pretreatment levels ranged from 7.9-12.2 ppm. After 10 days levels ranged from 1.9-5.5 ppm.

Different treatment rates had no significant effect on pH. In the summer, pH in all ponds was significantly lowered during the first 10 days after treatment. Before treatment pH ranged from 9.1-10.0. About 10 days after treatment the range was 8.1-9.8. Within 15 days after treatment, pH had risen, approaching or surpassing pretreatment levels. In winter, ammonia also significantly lowered pH through the first 15 days after treatment. Unlike summer, there was no trend towards recovery. Winter pretreatment pH ranged from 9.3-9.8. After 15 days the range was 8.4-8.8.

In summer, Treatment 1 had no effect on alkalinity. All other treatments caused significant increases. After 10 days alkalinity dropped and approached pretreatment levels. Alkalinity in ponds receiving the 3 higher applications ranged from 77-110 ppm before treatment, and 87-163 ppm about 10 days after treatment. In winter, all treatments increased alkalinity. Unlike summer, there was no trend towards recovery after 10 days. Alkalinity ranged from 95-120 ppm before treatment to from 130-178 ppm 15 days after treatment.

Ammonia applications did not significantly affect hardness in any study ponds. Summer readings ranged from 73-108 ppm before treatment to from 75-110 ppm about 15 days after treatment. Winter readings ranged from 108-125 ppm before, to from 105-165 ppm 15 days after treatment.

Conductivity data were not collected during all segments of this study. Therefore these data were not statistically analyzed. Observations indicated trends of increased conductivity after ammonia applications. Peak conductivity occurred about 5 days after summer treatment and about 7-19 days after winter treatment. After conductivity peaked, there was a trend toward normal levels. Conductivity before treatment ranged from 215-348 μ mhos/cm and from 242-440 μ mhos/cm 10 days after treatment.

Analysis of turbidity data was inconclusive. Both summer and winter results showed slight increases. Most values approached or fell below pretreatment levels within 10 days. Pretreatment turbidity for all seasons ranged from 1-9 JTU. Ten days after treatment the range was 5-28 JTU.

Ammonia had no effect on nitrate/nitrite nitrogen readings in the study ponds. Summer readings ranged from 0.9-3.2 ppm before, and from 2.0-3.2 ppm 15 days after application. Winter readings ranged from 2.1-3.6 ppm before, and from 1.3-4.0 ppm 15 days after treatment. Although some shifts occurred, no clear trend appeared.

Decay Rate Model

Development of a decay rate model to predict how long anhydrous ammonia remains in an ecosystem received considerable effort. Emphasis was placed on giving fishery managers a method of calculating decay or dissipation time for any selected NH₃ concentration at any water temperature. This model could also aid environmentalists who encounter ammonia spills.

All water quality data were combined giving 275 data points to develop the model. Using methods of stepwise multiple regression (Draper and Smith 1966), the three most important variables in determining decay rate were: 1) water temperature, 2) initial concentration, and 3) final concentration. Many combinations and transformations of these and other water quality parameters were tested. The small increase in precision obtained with some combinations (<5%) did not justify more complicated models. These three variables accounted for 75% of the total variation of predicted elapsed time. The predictive model is:

$$Y = a + b_1X_1 + b_2X_2 + b_3X_3$$

where:

Y = elapsed time to be predicted (days);

a = intercept (y-axis);

b₁ = regression coefficient of X₁;

X₁ = starting ammonia concentration (in management operations this is applied concentration, while in cases of pollution this is the ammonia concentration of the first sample at the point of the spill);

b₂ = regression coefficient of X₂;

X₂ = ammonia concentration after elapsed time (concentration accepted as safe);

b₃ = regression coefficient of X₃; and

X₃ = water temperature (F) (temperature at time of ammonia application).

Statistics for each independent variable are given in Table 2. Substituting these values and the calculated intercept (a = 23.755), our predictive model becomes:

$$Y = 23.8 + 0.602 X_1 - 0.777 X_2 - 0.251 X_3$$

The standard error of predicted elapsed time is ± 4 days, using the means for each independent variable listed in Table 2. Discretion should be used when estimating elapsed time from independent variables which fall outside of ranges used in this study.

Table 2. Means, ranges, and regression coefficients for independent variables used in the anhydrous ammonia decay rate model (elapsed time (days) = 23.8 + 0.602 X₁ - 0.777 X₂ - 0.251 X₃).

<i>Independent Variable</i>	<i>Mean</i>	<i>Range</i>	<i>Regression coefficient</i>
Starting Conc. (X ₁)	30 ppm	15-41	0.602**
Final Conc. (X ₂)	18 ppm	0.5-30	-0.777**
Temperature (X ₃)	68 (F)	47-85	-0.251**

**Significantly different from zero at the 0.01 level

At concentrations applied, ammonia remained toxic for approximately 21 days in summer and 30 days in winter tests.

Benthic Organisms

Benthic organism numbers were reduced significantly and to approximately the same degree by all ammonia applications except Treatment 1 in the winter of 1973 (Table 3). Numbers of benthic organisms decreased after summer applications and remained low at least 60 days, but recovery began after 33 days. Numbers of organisms dropped during the first 33 days after winter applications, but these data failed to indicate what happened thereafter.

Table 3. Mean number of benthic organisms observed per sq. ft. (0.09 m²) in ponds treated with anhydrous ammonia, Heart of the Hills Research Station 1971-73. Time periods are defined as: I, before ammonia applications; II, 1-33 days after application; III, 34-60 days after application.

Time Period	Treatments				Treatments			
	1	2	3	4	1	2	3	4
	Summer 1971				Summer 1972			
I	258	178	178	238	122	126	70	141
II	113	93	27	11	71	14	23	51
III	87	57	89	31	37	163	18	59
	Winter 1972				Winter 1973			
I	93	348	476	198	106	75	213	188
II	54	349	66	117	177	62	158	104
III	34	46	44	46	151	405	125	318

Aquatic Vegetation

Anhydrous ammonia significantly decreased vegetation cover in all study ponds (Table 4). Although ammonia appeared more effective in summer, any concentration above 15 ppm destroyed most vegetation observed, regardless of season.

Table 4. Mean percent vegetation observed in ponds treated with anhydrous ammonia, Heart of the Hills Research Station 1971-73. Time periods are defined as: I, before ammonia application; II, 1-33 days after application; III, 34-60 days after application.

Time Periods	Treatments				Treatments			
	1	2	3	4	1	2	3	4
	Summer 1971				Summer 1972			
I	60	98	87	71	92	87	77	97
II	8	5	8	2	6	12	3	1
III	37	45	45	35	6	7	15	50
	Winter 1972				Winter 1973			
I	98	75	87	89	98	95	70	95
II	86	56	66	78	65	1	22	0
III	45	8	2	15	16	0	0	0

Aquatic plants were abundant in study ponds before ammonia applications. Muskgrass (*Chara* sp.) and bushy pondweed (*Najas* sp.) were predominant plants in all ponds during summer observations. Pondweed (*Potamogeton* spp.) and water stargrass (*Heteranthera* sp.) were also present in ponds. Horned pondweed (*Zannichellia* sp.) was least abundant in most ponds during summers. Plants listed above were present in all ponds during winter observations, but *Zannichellia* was predominant.

Most masses of *Chara* had turned white within 24-48 hr after summer ammonia applications. Defoliation of aquatic plants occurred about 3 days after applications. The only plant which continued to survive to any extent was *Heteranthera*, which maintained a

green stem. A trend toward vegetation recovery appeared about 33 days after summer applications but percent cover remained significantly less than before applications.

Plant responses to winter ammonia applications were similar to responses observed in summer. Vegetation, except *Zannichellia*, was affected by all concentrations. *Zannichellia* was not completely destroyed by any concentration and seemed unaffected by 15 ppm ammonia. Vegetation cover was still decreasing up to 60 days after winter ammonia applications.

Bioassay

There was no significant difference in effects on fishes between summer and winter ammonia applications of Treatments 1, 2, or 3, but there was a significant difference in effects of Treatment 4 in the two seasons. Treatment 4 killed all fishes in a relatively short time, regardless of season. For consistency in analysis all seasons were combined giving one set of mortality readings for each concentration.

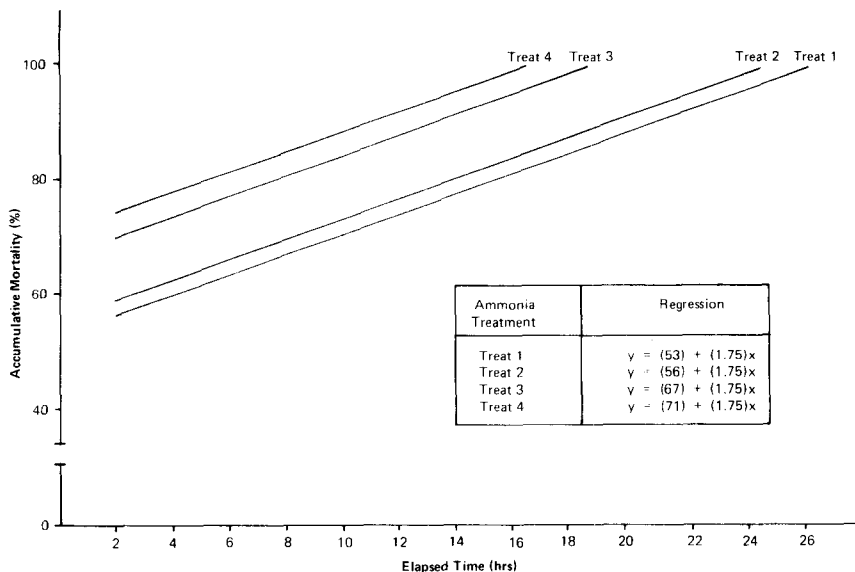


Figure 1. Accumulative Mortality—time relationship determined using all species studied in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

Accumulative mortalities of all species combined provided mortality regressions for each ammonia concentration (Figure 1). There were no differences in slopes of the 4 regressions, but highly significant differences in intercepts. A pooled slope was calculated and used in each regression. Effects of anhydrous ammonia on individual species were analysed (Table 5) and plotted in the same manner (Figures 2-7). All applied concentrations of ammonia caused high mortality within 27 hr. In most cases complete fish kills were not realized at concentrations less than 30 ppm.

There was a rapid reaction of fishes to all ammonia application rates. Reactions were faster and more pronounced with application of Treatments 2, 3, and 4. Young fish responded first, followed soon by adults. Affected fishes exhibited a frenzied erratic behavior near the surface. Many fishes affected in deep water underwent a spiraling motion towards the surface. It was not uncommon to see fishes jumping from pond to

shore. Usually fishes died quickly after surfacing. Fishes examined after death displayed reddened gills and fins.

Table 5. Analysis of covariance on regressions of accumulative mortality on elapsed time, testing effects of different concentrations on species stocked in ponds treated with anhydrous ammonia, Heart of the Hills Research Station, 1971-73.

Species	F-values when comparing		
	Slopes	Intercepts	Degrees of Freedom
Goldfish (<i>Carassius auratus</i>)	4.88**	7.28**	(3 and 40)
Black Bullhead (<i>Ictalurus melas</i>)	2.93**	6.67**	(3 and 40)
Channel Catfish (<i>Ictalurus punctatus</i>)	1.48	2.27	(3 and 40)
Green Sunfish (<i>Lepomis cyanellus</i>)	2.19	4.60**	(3 and 40)
Bluegill Sunfish (<i>Lepomis macrochirus</i>)	1.69	3.06*	(3 and 40)
Largemouth Bass (<i>Micropterus salmoides</i>)	0.72	3.98*	(3 and 30)

* (P<.05)

** (P<.01)

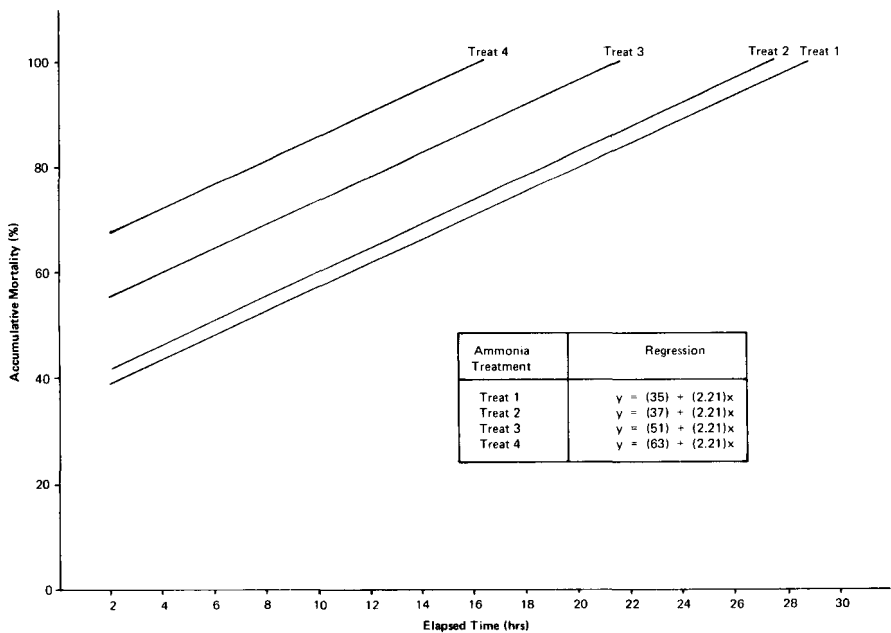


Figure 2. Accumulative Mortality—time relationships for goldfish (*Carassius auratus*) in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

DISCUSSION

Anhydrous ammonia holds promise as a fishery management tool in its combined role as a fish toxicant, aquatic herbicide, and fertilizer. Ammonia is a naturally occurring substance throughout the environment, and its application to a pond adds no foreign substance to the water.

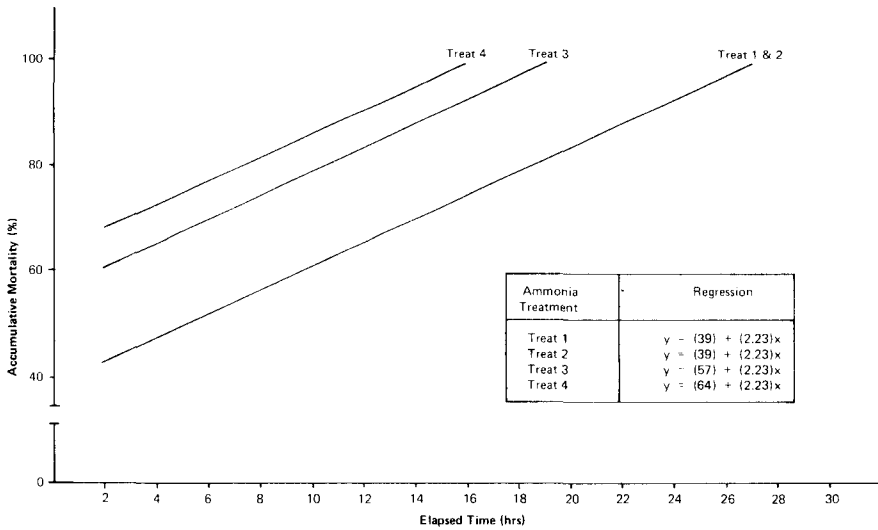


Figure 3. Accumulative Mortality–time relationships for black bullhead (*Ictalurus melas*) in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

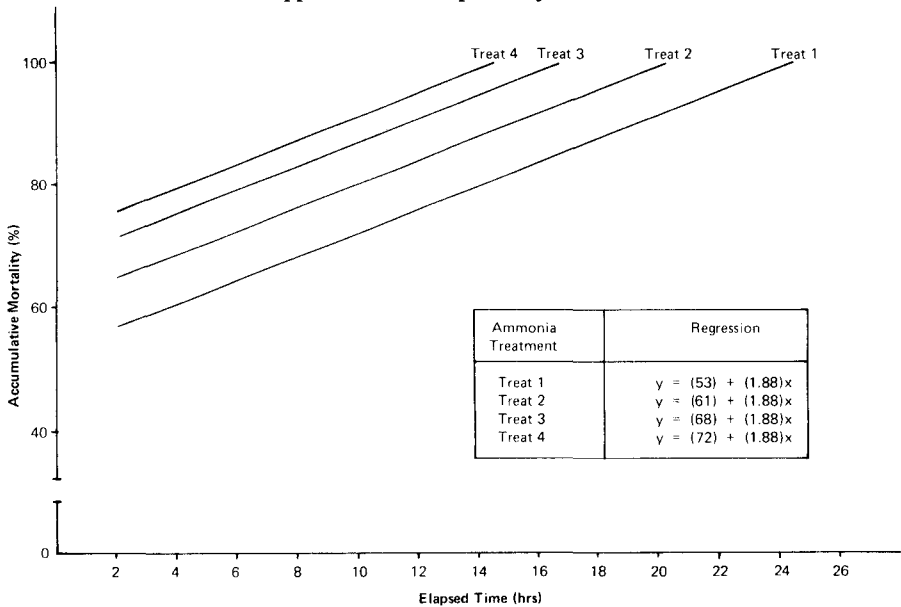


Figure 4. Accumulative Mortality–time relationships for channel catfish (*Ictalurus punctatus*) in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

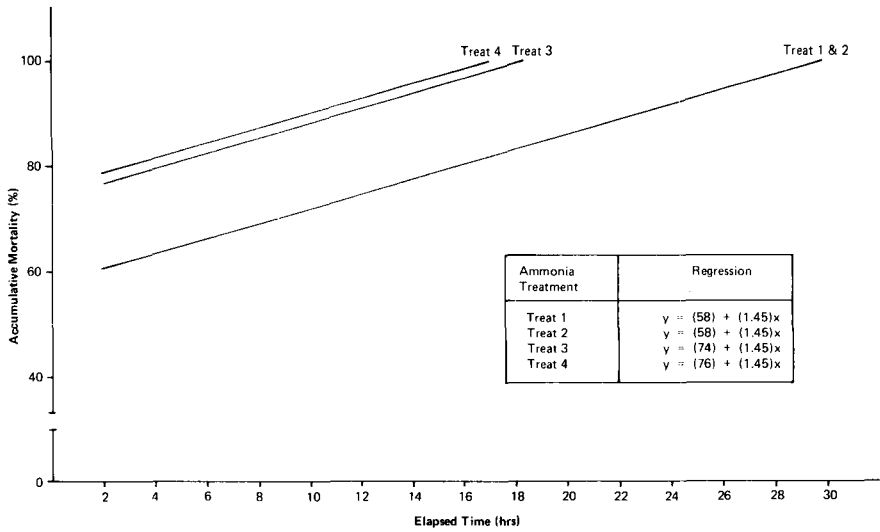


Figure 5. Accumulative Mortality—time relationships for green sunfish (*Lepomis cyanellus*) in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

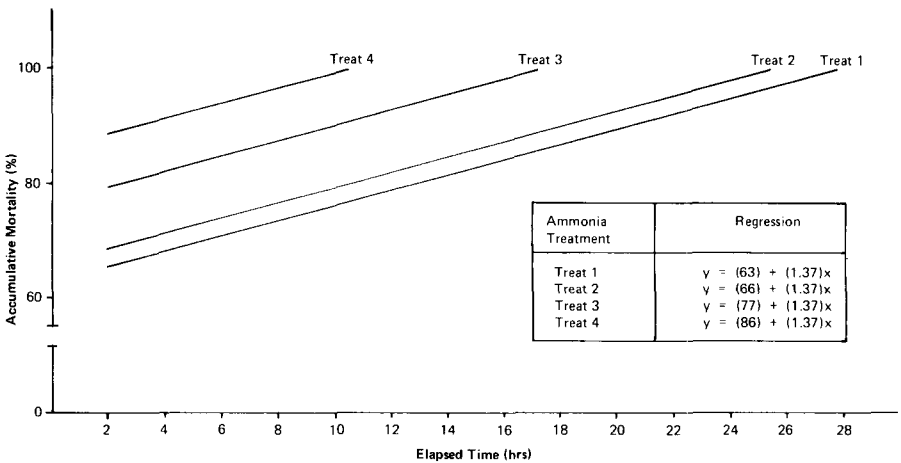


Figure 6. Accumulative Mortality—time relationships for bluegill sunfish (*Lepomis macrochirus*) in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

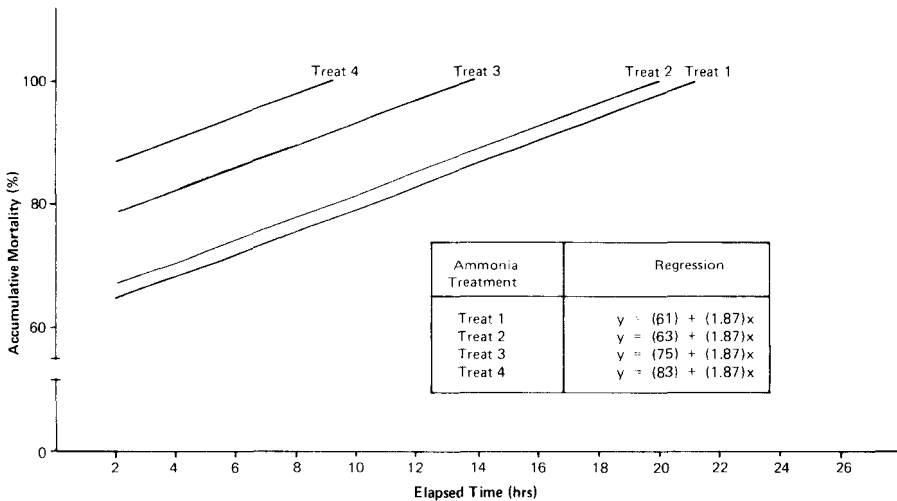


Figure 7. Accumulative Mortality—time relationships for northern largemouth bass (*Micropterus salmoides, salmoides*) in ponds treated with different ammonia concentrations, Heart of the Hills Research Station, 1971-1973. Treatments 1, 2, 3 and 4 approximate 17.8, 26.3, 32.9 and 39.3 ppm ammonia respectively.

Additions of anhydrous ammonia, at concentrations above 15 ppm, had profound effects on pond ecosystems. Ammonia remained toxic for varying amounts of time, depending on initial concentration, water temperature and other factors. Decay or dissipation time can be predicted, using the model given.

Although significant decreases in pH were observed, pH values remained high. Changes were probably held to a minimum by the buffering complex of the water. In weakly buffered waters, addition of ammonia would probably increase pH (Brockway 1950; Jones 1957; McKee and Wolf 1963; and Trussell 1972). Dissolved oxygen levels also decreased quickly after ammonia applications. This decrease was in response to increased B.O.D. caused by decomposition of plants and animals killed by ammonia.

Increases were observed in alkalinity, conductivity, and turbidity. Turbidity increases were believed correlated with the pulses of phytoplankton production following applications. No changes were seen in hardness or nitrate/nitrite nitrogen after ammonia applications. Results of the nitrate/nitrite nitrogen analysis seem to indicate ammonia was not cycling through the system in appreciable amounts. Analysis of pond bottom soils in 1975 showed no increase in ammonia or nitrates (Klussmann 1973). It is believed that introduced ammonia was lost to the atmosphere.

Benthic organisms were not completely removed from any pond by ammonia. Application rates over 15 ppm should, however, significantly decrease populations of benthic organisms.

Anhydrous ammonia as an aquatic herbicide is promising. Although the chemical seems more effective in summer, any concentration above 15 ppm will kill most vegetation, regardless of season.

Anhydrous ammonia, at the lowest application rate, effected high mortalities of all fish species observed. Complete fish kills, however, are not likely at concentrations less than 30 ppm. Greater concentrations may be necessary in water with low pH due to shift of the equilibrium reaction of ammonia with water. Rough fish species were almost as susceptible to the toxin as game fish species, but goldfish and black bullhead appeared to be somewhat more tolerant of ammonia than other species observed. Ammonia had nearly equal toxicity

in both summer and winter, allowing a wider range of application than other toxicants which are effective only in narrow temperature ranges.

Anhydrous ammonia is less expensive than other currently used fish toxicants and aquatic herbicides. Current cost of anhydrous ammonia is about \$0.10 per 454 g (1 lb). A 30 ppm application would cost \$8.10 per 1,292 m³ (acre-foot). Much of the equipment needed to apply ammonia is available from ammonia dealers. Gaseous ammonia is extremely dangerous, so safety procedures must be observed.

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