

Figure 2. Comparison of survival of young oysters in two areas of different salinity in Mobile Bay.

A CASE OF GUANOTROPHY IN A SMALL LIMESTONE QUARRY POND IN KENTUCKY *

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INTRODUCTION

The word "guano" comes from the Peruvian Spanish word "huanu" meaning dung, and it was used originally to refer to the massive bird droppings on some of the offshore islands near Peru. These droppings were found to be a good fertilizer source, as they were high in nitrogen and phosphorous. The term "guanotrophy" was perhaps first used by Leentvaar (1958), and it has to do with the producing of a specific environment by bird excrements. (Dr. Brinkhurst introduced the term to some of us at the Midwest meeting last year when he described guanotrophy in a British lake).

In a nation and in a world where poultry production is becoming an ever increasing source of protein for human consumption, the disposal of fowl wastes in such a manner as to not pollute the environment presents a great challenge to scientists in many disciplines; and, here in the beginning we raise the question, "Can any good come out of fowl wastes?"

^{*} Most of this paper was first presented at the annual meeting of the Midwest Benthological Society in 1967.

Our interest in droppings has increased greatly during the last 12 months; but, in order to get the full story, it will be necessary to go back seven years.

EARLY FISHERY MANAGEMENT OF ALLEN POND

A one-fourth acre limestone quarry pond in Kentucky was started on a fishery management program in 1959, and 3 to 5 domestic mallards were reluctantly allowed to live on it. No fishing was allowed in 1959. In the period 1960 to 1963 this quarry had been fertilized with inorganic fertilizer during the growing seasons and had produced annual catches of bluegills and largemouth bass of 128, 121, 94 and 33 pounds respectively (Howell, 1965). On a per acre basis this would be catches of 512, 484, 376, and 132 pounds. In 1963 the pond was out of balance and the fish were poisoned; the pond was restocked in April, 1964, for another experiment in which a combination of bluegill, shellcrackers and largemouth was used.

No fishing was allowed in 1964. In the middle of June, 1964, the pond turned an unusual dark brown, and there were streaks of flocculent material which were assumed from the bank to be nothing but rather strange microcrustacean pulses. Fertilizer was applied on June 20, and on June 24 and 25, forty-two dead fish were picked up. Later applications that year resulted in no fish mortality.

In 1965 seventy-three pounds of bass and bluegills were caught. Fifteen bream died in mid-April during the spring turnover; there were no other deaths during the growing season. For most of the summer the water had a brownish appearance, and fertilizer applications only resulted in short-lived phytoplankton blooms. The best fisherman in the neighborhood also remarked that the fish were not in as good condition as for the first fishing experiment. Cool fall nights brought about a turnover which lifted brownish, black flocculent material to the surface, and for the first time it was more than suspicioned that duck droppings were the cause.

EVENTS BEFORE AND AFTER THE TOTAL FISH KILL IN SPRING OF 1966

Fertilizer applications were started as usual on March 15. The water was clear at this time. The second application was made one week later, and the water began to become slightly murky. A third application was made on April 7, for the water looked a rather sickly brown. The ducks had to go. Two were spirited away on the night of April 9. The other three escaped to the pond. On April 13 the owner left for the meeting of the Midwest Benthological Society in Mt. Pleasant, Michigan. When he returned on April 16, all fish were dead.

Family members stated that on the morning of April 14 the water in the pond had a milky appearance. By noon it had turned black, and between 1:30 and 2:00 pm the first dead fish were noted. A strong, indescribable odor was coming from the pond. Fish continued to die on Friday. When the owner returned on Saturday, the water was still black, and a floating black-brown cloud of flocculent material seemed to be moving up and down in the water. There were only 10 fish floating on the water at this time, and they were removed. The other three ducks were caught and taken to the country.

Fish continued to come to the surface for another ten days, with most of them coming up on Saturday, April 23. A total of 148 bream and 7 bass were picked up. The largest bream was a shellcracker that weighed 12 ounces. There was only a handful of fingerling bream, but it is likely that the ducks ate some that were dying. The 7 bass weighed from 10 to 16 ounces each, indicating that forage fish had been inadequate during the two growing seasons they had lived. In the first experiment the annual weight increase had been 10-12 ounces.

EFFORTS TO READY THE WATER FOR MORE FISH

During the period April 16-23, the dissolved oxygen had ranged from 9 to 16 ppm, and the pH from 9.5 to 10.0. On April 25 a blunt-nosed minnow and a sunfish were placed in 3 gallons of pond water. Twelve hours later the minnow gasped briefly at the surface and died. The sunfish was found dead on the bottom 13 hours later. On April 27 two legallon jars were filled two-thirds full of pond water and one bluegill was placed in each. One of these jars was aerated. A fish in tap water served as the control. Again, the fish in the pond water died overnight. The fish in the aerated water had jumped out. The control fish was alive. The next night the experiment was repeated, except one fish was placed in the jar which had been aerated the previous night. By morning two fish were still living, but the one in the unaerated pond water was dead. The other two continued to live.

Frankfort fishery biologists were contacted, and it was suggested that hydrogen sulphide might be the culprit. The college chemistry department's lead acetate papers proved unsuccessful in detecting any dissolved H_2S . Some bottom sludge was placed under a hood and HC1 was poured over it. A strong rotten-egg odor was given off. A Hach Chemical Company hydrogen sulfide kit arrived on May 10, and three samples were checked for the dissolved gas. The samples were taken from 18, 36, and 54-inch depths. All three tests were negative. A 10 cc plankton tow vial which had been placed in the kitchen window April 16, was shaken and filtered. The filtrate was quickly tested and was found to have 0.1 ppm of hydrogen sulfide.

On May 16 a gallon jar was filled one-third full by hand with bottom muck. A water sample was also taken 18 inches above the muck and found to have a trace of sulfide. When the flocculent material in the jar had settled, a sample was poured off the top and tested for sulfide; a positive test of 0.05 ppm was obtained. The jar was then shaken, a sample filtered and using a Hach Wildlife Kit the following analyses quickly tested; 3.5 ppm were present. From a water sample taken at a foot depth, the D.O. was found to be 13 ppm; the carbon dioxide, O.O. In 5 feet of water just beneath a duck roost 0.45 ppm of H₂S was found.

TABLE 1. Partial water analysis of Allen	Pond, May 16, 1966
Carbon dioxide Dissolved oxygen Phenolphthalein alkalinity (high range) Methyl orange alkalinity (high range) pH	
Temperature (Centigrade)	
0.5 feet	
3.0 feet	18.0
5.0 feet	

On May 18 a 7.5 horsepower outboard motor was used to zig-zag a boat through the pond for 20 minutes. Twenty minutes after the aeration, three sulfide determinations were made. One collecting site was below the same duck roost mentioned above. In all instances only traces of the gas were found. When samples were run 1½ hours later, no gas could be detected. The water temperature no longer had the 5° spread of two days earlier; it was 20.0° near the surface and 19.5° near the bottom.

The pond was now considered ready for a try at stocking. Two fisherman friends caught 63 large fingerling bluegills on May 20, and placed them in the pond. They were still alive on May 23, and a minnow seine was used to secure an additional 190 fingerling which would bring the total to the desired 250 fish. On May 25 dead fish began to come to the surface. A H_2S test taken 6 inches above the bottom showed no measurable gas; however, many of the dead fish had their mouths filled with black organic matter, indicating that they might have been feeding on the bottom when overcome.

Douderoff and Katz (1950) mentioned that the lower the pH of the water the more readily H_2S would become toxic and also that possibility high pH's could produce lethality. In the evenings the pH had been running from 9.8 to 10.2, and several possibilities were considered whereby the pH might be lowered, among them were the adding of muriatic acid or NH₄SO4. Frankfort suggested that the ammonium sulphate be tried. Eighty pounds of the sulfate was broadcasted on the pond on June 13. By the next evening the pH was 10.5 and the H₂S above the bottom was 0.5 ppm. An air pump was secured on June 18, and the aeration of the pond was started, and over a 10-day period the water was aerated 4 to 5 hours per day. Friends began restocking process on June 28. Out of the first 150 bluegills, 30 died. Stocking was completed on July 19. No fish died after July 1 and the pond seemed in excellent condition for the rest of the growing season. Plankton and bottom organism growth was vigorous, and excellent fish hatches occurred in August and September.

GUANO ANALYSIS AND THE SULFUR CYCLE

The exact extent to which hydrogen sulfide evolvement can be tied in with guanotrophy cannot be determined easily for there are so many other factors entering into the biogeochemical cycles of a pond ecosystem. In the early years of the fishery management program when there were 3 to 4 inches of decaying duckweed in the bottom, the fertilization program apparently resulted in no serious hydrogen sulfide evolvement. In Allen Pond the trouble seemed to begin with the buildup of the duck fecal material. What in the manure could cause this? No complete analysis of duck guano could be found in the literature. Sanderson (1953), in studying the duck droppings from Long Island, New York, duck farms, found that 1,000 ducks left behind 96 pounds of suspended solids per day of which 58 pounds were volitile; the total nitrogen was 5.7 pounds, the total phosphate 7.6 pounds and the soluble phosphate 3.6 pounds. These figures are lower than Copeman and Dillman (1937) found when analyzing the excrements of sea birds on the breeding grounds in the Union of South Africa during the period 1921-1928. Their average analytical results were 10.0 per cent total nitrogen, 11.2 per cent phosphoric oxide and 2.0 per cent potash. No mention was made by any of these authors about sulfur compound residues. No doubt, some sulfur can leave the ducks in the form of the three sulfur-containing amino acids which are excreted by the kidneys. Ryther (1954) even found uric acid decomposing bacteria associated with duck farm effluents.

Hutchinson (1957) and Morgan and Lackey (1965) discuss at length the sulfur cycle in fresh waters. These authors bring out that probably more hydrogen sulfide is evolved by bacteria under anaerobic conditions than aerobic conditions. Members of the genus *Desulfovibrio* are the best known bacteria which reduce sulfate to hydrogen sulfide. This gas can also be produced in the bacterial decay of any organic matter which contained reduced sulfur. Sulfite contains reduced sulfur. Sulfite, hyposulfite, thiosulfate and elementary sulfur can be reduced as well as sulfate; bacteria can also oxidize these same compounds.

From a bottom sludge sample taken in February, 1967, for bacterial culture in the laboratory both aerobic and anerobic hydrogen sulfide evolving bacteria were found to be present. The anaerobes gave a much more positive test for the gas. An aerobe, *Thiodendron*, had tentatively been identified in association with the yellow flocculent material collected with a plankton sample taken in early November, 1966. Morgan and Lackey reported 29 species of sulfur bacteria from Warm Mineral Spring, Florida. To describe the natural ecological community of these bacteria, he used the term "Sulphuretum," which had been earlier used by Bass-Becking. The Florida spring had 1,704 ppm of sulfate.

HYDROGEN SULFIDE LETHALITY

Hydrogen sulfide has been found to be lethal in small quantities in the air and dissolved in water. Turner (1965) states that in concentrations above 0.05 per cent the gas is lethal in closed polltry houses, and that it is tolerable only when it is below 0.004 per cent. Morgan and Lackey (1965), in the anaerobic warm mineral spring which they studied, found that sulfide bacteria thrived until the sulfide concentration was less than 0.085 mg/1; and that *Thiodendron* in the lab could tolerate sulfide concentrations up to about 1.5 mg/l. There were also over 150 species of animals found in this spring where the average hydrogen sulfide concentration was 0.240 ppm. The Metazoa found were usually dwarfed or deformed.

Douderoff and Katz (1950) reviewed critically the literature dealing with the toxicity of hydrogen sulfide on fish. They cited 19 references and were forced to conclude that most of them could not be effectively evaluated and interpreted because of insufficient data. Douderoff and Katz concluded that hydrogen sulfide can be toxic to some fresh-water fish in concentrations well below 1.0 ppm.

Bonn and Follis (1967) studied the effect of total sulfides on catfish and bluegill in the laboratory. In 3-hour test periods the quantity of dissolved gas necessary to produce a TL_m of the test catfish varied from 1.82 to approximately 7.0 ppm, depending upon the pH of the water. The higher the pH, the more of the sulfide was necessary. At a pH of 7.0 the TL_m of unionized hydrogen sulfide was 1.0 ppm for fingerling channel catfish, 1.3 for advanced fingerlings and 1.4 for adult catfish. Bluegill were much more tolerant; only 2 fingerlings died during the 3-hour exposure to a 1.6 ppm unionized hydrogen sulfide exposure. These same workers found that in the field in acid lakes in Texas, maximum production of the gas can be expected April through June; and this furnished them with a possible explanation as to why only a few fry and fingerling bluegills and catfish were ever found. This could also be true in alkaline Allen Pond—there seemed to be less of a successful hatch each year of increasing guanotrophy. (It should be remembered that fish lethality continued from April 14 until the middle of June when aeration was started).

The April 14 fish kill seemed to kill some of the bottom organisms also. No chironomid emergings were observed during the next two or three weeks. A large crop of Chaoborinae larvae were swimming oddly near the surface on April 17, but all appeared to be dead on the 18th. Corixids and adult gyrinids continued to live.

A 5-pound snapping turtle came to the surface on September 28; its body was badly decomposed and covered with moss (Algae).

DISSOLVED OXYGEN AND pH

In the years previous to 1965 when a Hach Wildlife Kit, AL-36-P, was obtained, pH field tapes and the chemistry laboratory pH meter were used to determine pH. Determinations were made only two or three times during the growing season, and in each instance the pH ranged from 9.0 to 9.5. No oxygen determinations were made prior to the securing of the kit. When the kit was first used in April, 1965, the pH was 9.6 and the dissolved oxygen 10 ppm at a 6-inch depth. Later that season the D.O. went to 12 to 18 ppm.

After the fish kill in April, 1966, rather frequent dissolved oxygen and pH determinations were made at a 6-inch depth at 7 pm. These

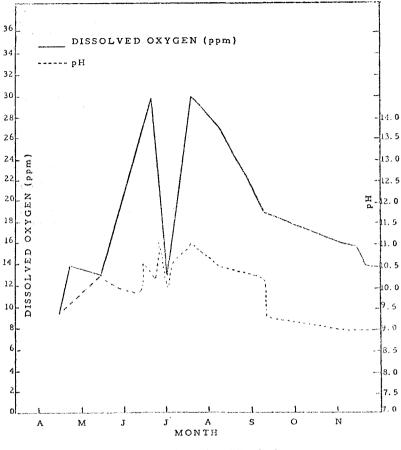


Figure 1. Fluctuation of dissolved oxygen and pll, April through November, 1966

determinations are plotted on Figure 1. Occasional samples were taken 6 inches above the bottom early in the morning and in the evening. At the 6-inch depth in the evening the pH ranged from 9.0 to 11.0; the oxygen from 10 to 30 ppm. The two sharp dips in pH are synchronized with the aeration by outboard motor on May 18 and with the aeration with garden hose during the last few days of June. The pH's of 11.0 occurred after the addition of the ammonium sulfate and the ensuing phytoplankton buildup. On July 18, when the dissolved oxygen was 30 and the pH 11.0 at the 6-inch depth, 6 inches above the bottom the D.O. was 7 ppm and the pH 9.2. On August 8 the dissolved oxygen increased from 7 ppm in the morning at 6 inches to 27 ppm in the evening; the pH was 9.0 in the a.m. and 10.5 in the p.m. On that same date at 6 inches above the bottom in the morning the dissolved oxygen was 7 ppm and in the evening it was 11 ppm; the pH changed from 8.8 to 9.1.

Douderoff and Katz (1950) concluded after reviewing the literature that optimum pH range for fish was 5.0 to 9.0, and that extremes of 4.0 and 10.0 could be tolerated.

In Allen Pond there was no evidence that high pH's or DO's brought about the death of the fish, even though the owner thought for a while that they might be playing a role in prolonging the time of toxicity.

THE DUCKS AND THE GUANOTROPHY

Over the 7-year period of fishery management of Allen Pond, 3 to 5 ducks had been on the pond, with the exception of the second and third growing seasons when they were removed to prevent dabbling for fish eggs. During this time it is estimated that there were 9,544 duck days spent on the pond, with an average of 3.7 ducks/day/year. Using the evidence gathered by Sanderson (1953) on duck feees and the estimated that the ducks on Allen Pond added about 0.06 pound/day of suspended solids to the water. If the ducks were on the water 80 per cent of the time this would mean 458 pounds of solids were added before the ducks were removed, and in this material there would be 26.1 pounds of nitrate and 34.8 pounds of total phosphate. On a per acre basis this would be the equivalent of 1,832 pounds of solids, 104.4 pounds of nitrate and 139.2 pounds of total phosphate being added.

It is hypothesized that the use of inorganic fertilizer probably speeded up guanotrophism.

Since there is relatively no flushing of water in the quarry, there was a maximum amount of fecal accumulation, and the sludge gradually sealed over the slowly decaying leaves and duckweed, promoting anaerobic conditions within the bottom material.

Evidences of guanotrophy in this eutrophic habitat seemed to be:

1. the sealing of the bottom and the establishment of anaerobic conditions within bottom muck

2. the establishment of a sulphur bacterial cycle

3. the evolvement of hydrogen sulfide gas

4. the lifting of flocculent, sludge-like material

5. a decrease in the number of photosynthetic *Euglena* and colonial flagellates, and an increase in the pigmentless Euglena and in *Chlamydomonas*

6. a decrease in the number of diatoms and microcrustacea, and an increase in the number of ciliates and rotifers

7. a decrease in the number of successful fish hatches

Leentvaar (unpublished paper) found that in a lake in the Netherlands where guanotrophy was increasing, the dominant plankton organism, *Dinobryon pediforme* (flagellate), was gradually replaced by *Chlamydomonas*, and that the number of *Euglena*, *Lepocinclis* and *Mallomonas* also increased; the number of *E. coli* seemed to fluctuate with the seasonal changes in the number of birds. Morgan and Lakey (1965) observed that ciliates constituted the largest group of species in the sulfuretum in Florida; they listed 98 species, and most of them were from the bottom material, and all seemed to be able to thrive in the presence of some hydrogen sulfide. The ciliates became most noticeable in Allen Pond as the fall turnover dispersed flocculent material from the bottom throughout the pond. The June aeration seemed to eclipse the *Chlamydomonas* buildup.

WHAT IS GOING TO HAPPEN THIS SPRING

There are still 2 to 3 inches of sludge on the bottom of the pond. It has already been mentioned that sulfide evolving bacteria were cultured in the laboratory from loops of muck material collected February 18, 1967. On March 15 a water analysis was run with the help of one of the state fishery biologists. The results of this analysis are presented in Table 2.

TABLE 2.	Physico-chemical	characteristics	of	Allen	Pond,
	March 15, 1967				

Temperature	15° Centigrade
Dissolved oxygen	13.00 ppm
Carbon dioxide t	race
Alkalinity (phenolphthalein)	13.68 ppm CaCO ₈
Alkalinity (methyl orange)	32.08 ppm CaCO ₈
Hardness	46.90 ppm CaCO ₈
Total Iron	0.60 ppm
Total phosphate	0.87 ppm
Total sulfate	98.00 ppm
Ammonium nitrogen	0.50 ppm
Total nitrate	2.12 ppm
pH	8.7

The ingredients are there for another fish kill, but the signs do not point in that direction now. The water is cloudy with a real good plankton bloom for this time of the year. Last year the bottom could be seen all over. The ducks are not there. No fertilizer has been applied. The pond does not look sick.

The question was asked in the beginning, "Can any good come out of fowl wastes?" The answer now might be, "Yes, if we can get another year of free fertilization simply by blowing a little air through the bottom sediments that would be fine."

"What if the fish die?"

"Never let me see another duck."

ADDENDUM

WHAT HAS BEEN LEARNED SINCE MARCH 15, 1967?

1. On April 26, 1967, five bluegills came to the surface; 20 more surfaced in period May 29 to June 15; and on July 21, about 30 hours after fertilizing the pond, 19 bluegills and one large-mouth bass surfaced. There were no further kills during the year. The pond was aerated on May 29 and 30, and on June 27. The dead fish weighed 4 to 10 ounces. (A few fingerling may have been overlooked.)

2. Total sulfide was determined at two stations 6 inches above the bottom muck on the July 21 date; at one there was 0.3 ppm, and at the other a trace.

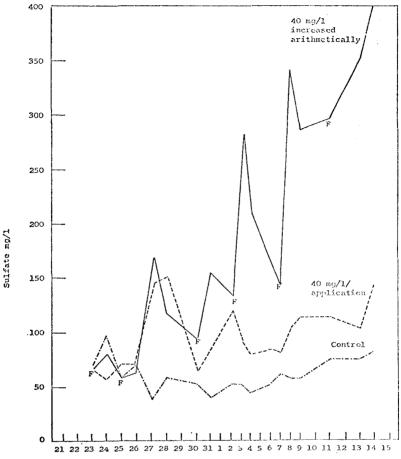
3. There have been no fish kills since 1967.

4. The fish catch in 1967 was 74 bluegills (44 dead and 30 living) with an average weight of 7.4 ounces, and a total weight of 34 pounds. An additional 174 small bluegills about 1.75 ounces each were thrown over the fence. This made a total removal for the year of 53 pounds of bluegill. The highest poundage in the 10-year history of the pond was caught in 1968; there were 606 edible bluegill removed having a total weight of 133 pounds.

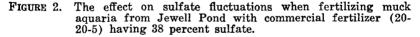
5. With muck and water from three quarries, three series of experiments using 16 muck aquaria in which there were 2 liters of muck and 6 liters of water, it was demonstrated that,

a. sulfide bacteria do not normally occur in the water above the muck.

b. sulfide forming anaerobes were present in the muck of all quarries. In the muck from the quarry where the ducks had been, the gas could be detected sooner than from the other two quarries. In the guano aquaria in 10 days it was 0.50 ppm, and in 15 days 3.00 ppm. In the nonguano aquaria it was 0.00 and 0.05 ppm. respectively.





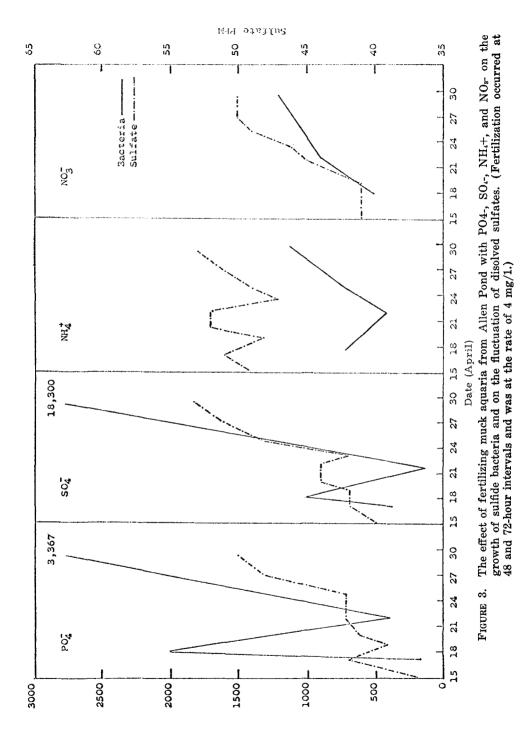


c. by using commercial fish pond fertilizer having 38 percent sulfate, the ability of the sulfide anaerobes to reduce sulfates was clearly demonstrated. In one aquaria the sulfate concentration aws reduced 135 ppm in 4 days (Figure 2).

d. by using a modified procedure of the Hach Lead Acetate Hydrogen Sulfide Test, 3.0 to 5.0+ ppm of total sulfide could be found in the muck of each aquaria at the end of an experiment.

e. in experimenting with four fertilizers (ammonium chloride, sodium sulfate, phosphoric acid and sodium nitrate) over a 15-day period, the phosphate increased the number of sulfide bacteria per ml³ three times as much as did the ammonia and nitrate; the sulfate increased the number 18 times as much (Figure 3).

6. During the first series of experiments in November, 1967, a special aquarium having two large fingerling bluegill, 10 liters of water and 2 inches of muck, was set up and allowed to evolve 5.0+ ppm of total sulfide in the muck; and then the muck was stirred to see if the fish would die. They both died within 5 minutes.



7. In the summer of 1968 the muck of 14 ponds in Kentucky was checked for total sulfide. Nine of the ponds were in the Inner Bluegrass; three of them were quarries and six were farm ponds. Sulfide was found to be present in all of the ponds with the exception of a recently constructed farm pond. In the quarries in August the total sulfide was usually 4.0 to 5.0 ppm. In the farm ponds the total sulfide seemed to vary with the amount of organic matter present in the muck, and only occasionally was a sample taken in which there were 5.0 ppm. The other five ponds sampled were in the Pennyroyal Region in Edmonson and Grayson counties, and the data was very similar to that found in the Bluegrass.

8. In the winter of 1969 a 31-gallon turnover tank was constructed in the laboratory to ascertain if a sudden temperature-produced water turnover could lift hydrogen sulfide and muck from the bottom. In two trial runs in which there was a 10° C. drop in surface temperature, the total sulfide in the top 6 inches changed from 0.00 to 0.06 and 0.08 ppm. No muck was lifted, and only 2.5 ppm were in it at the beginning. (Several additional runs are planned in the near future, when it is hoped to have nearer 5.0 ppm in the muck.)

9. What about guanotrophy? Three of the 14 ponds visited in 1968 had ducks. They all looked sort sick—the ponds, not the ducks.

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A SURVEY OF THE COMMERCIAL FISHERY ON FOUR OKLAHOMA RESERVOIRS

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ABSTRACT

The commercial fishery on four Oklahoma lakes (Eufaula, Gibson, Grand and Texoma) from which approximately 85 percent of the total state commercial harvest is landed was studied from July 1967 through June 1968. Thirty to forty-eight fishermen fished gill and trammel nets throughout the study period. Legal restrictions limited gear to 3 inch and larger bar mesh. The amount of fishing effort expended by mesh size and lakes was studied. Approximately 70 percent of the total effort was fished with 3 and 3½ inch bar-mesh nets. On the lakes studied, approximately 50 percent of the effort was fished on Lake Texoma. Monthly and yearly percent catch composition was determined and the average lengths, weights and condition factors for the fish harvested were computed. The catch was primarily composed of buffalo, flathead catfish, and carp with average weights of individual fish landed being 5.3, 5.0, and 7.5 pounds, respectively. During some months in various lakes a noticeable portion of the catch was carpsucker, paddlefish and channel catfish. The latter species could not legally be harvested. The best estimate of the harvest during the sampling year was 1,360,650 pounds on the lakes studies and 1,625,637 pounds for the total Oklahoma fishery. The average catch per 24 hours for 100 feet of net fished was 4.4 pounds. Lakes in northern Oklahoma yielded approximately 3 pounds per acre and Lake Texoma in southern Oklahoma yielded approximately 9 pounds per acre which was six percent of the standing crop other than clupeids.

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² Common names correspond to those in-,. A list of common and scientific names of fishes