

8-1933

A Study of the Tolerance of Certain Species of Fishes for Low Dissolved Oxygen & Increased Carbon Dioxide Concentrations

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1933

A STUDY OF THE TOLERANCE OF CERTAIN SPECIES OF FISHES FOR LOW
DISSOLVED OXYGEN AND INCREASED CARBON DIOXIDE CONCENTRATIONS.

BY

CHARLES R. ALLEN

A THESIS
SUBMITTED IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF
MASTER OF ARTS

WESTERN KENTUCKY STATE TEACHERS COLLEGE

AUGUST, 1933

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INTRODUCTION

Extensive investigations have been made of the oxygen and carbon dioxide content of water in which fishes live. A review of the literature, however, shows that these studies have not been made under controlled laboratory conditions dealing with the species studied in this investigation. It was the purpose of this study to determine the lowest concentration of dissolved oxygen that will support certain species of fishes and, also, the lethal effects resulting from increased carbon dioxide tension as determined under carefully controlled laboratory conditions.

REVIEW OF THE LITERATURE

Juday and Wagner (¹'08) placed perch (*Perca flavescens*) and croppies (*Pomoxis*) in aquaria containing water pumped from the depths of Lake Mendota free from dissolved oxygen. They found that both species died very soon; the perch, however, lived a little longer than the croppies.

Smith (²'25) lowered fishes in wire baskets to various depths in a moderately deep lake with a thermocline. The dissolved oxygen was determined for the various positions and asphyxiation points were assigned to the depths at which death occurred. The results of this experiment are similar to those obtained by Thompson during his observations in the Illinois River.

¹Juday, Chancey, and Wagner, George. "Dissolved Oxygen as a Factor in the Distribution of Fishes," Wisconsin Academy of Sciences, Arts, and Letters, Vol. XVI, Part 1, 1908, pp. 17-22.

²Smith, Frank. "Variations in the Maximum Depth at which Fish Can Live During Summer in a Moderately Deep Lake with a Thermocline," Bulletin Bureau of Fisheries, Document 970, 1925.

³
Thompson ('25) made studies of the dissolved oxygen concentrations in the Illinois River at points where fishes were being caught, where they had been taken but were absent at the time, and where fishes were dying. The conclusion was reached that dissolved oxygen concentrations between zero and two parts per million will not sustain life in any species of fishes. Other factors affecting the survival of fishes, however, were not eliminated.

⁴
Gutsell ('29) investigated certain aspects of water chemistry in relation to trout. Spring water in which no trout could live during the summer season tested low in dissolved oxygen content at that time. Fishes were tested in various mixtures of the spring water and water from a trout stream. The conclusions were reached that water containing between 1.3 and 2.5 p.p.m. dissolved oxygen would not sustain trout; that when the carbon dioxide content was increased to 39 p.p.m., the ill effects of reduced oxygen were not increased; and that trout are hardy to a considerable range of pH.

⁵
Weigilt ('85) reported that a large trout showed violent symptoms of distress when placed in water at 8°C containing 100 p.p.m. carbon dioxide. Another large trout showed signs of immediate distress when placed in water at 9°C containing 75 p.p.m.; however it later became quiet. Weigilt ('03) reported a later experiment in which a trout lay definitely quiet on its side in water at 10°C containing 1,000 p.p.m. carbon dioxide after one and one-half minutes. (It recovered in running water.)

³
Thompson, David H. "Some Observations on the Oxygen Requirements of Fishes in the Illinois River," State of Illinois Department of Registration and Education, Division of Natural History Survey, Bulletin, Vol.XV, Art.7, 1925.

4 & 5

Gutsell, James S. "Influence of Certain Water Conditions, Especially Dissolved Gases, on Trout," Ecology, Vol.X, 1929, pp.77-96.

Winterstein (⁶'08) states that fishes are sensitive to carbon dioxide tension rather than carbon dioxide content. At 20°C and with 5.85 p.p.m. dissolved oxygen, he found the carbon dioxide tension at the asphyxiation point to be 140 to 150 p.p.m. free carbon dioxide for *Perca* and 500 p.p.m. for *Carassius* (goldfish).

Reuss (⁷'10) found that in water at 10°C and well supplied with oxygen, the respiration of rainbow trout became dyspnoeic at a carbon dioxide concentration of 30 p.p.m.; that erratic movements were induced at 50, 63, and 83 p.p.m.; and that at 88 and 107 p.p.m. the trout turned on their backs.

Shelford and Allee (⁸'13) found that *Ameiurus* was narcotized in water containing about 320 p.p.m. carbon dioxide and 1.43 p.p.m. oxygen while other fishes died in this medium.

Wells (⁹'13) states that the combination of high carbon dioxide and low oxygen was more readily fatal to the coarse fishes used than low carbon dioxide and low oxygen.

Shelford (¹⁰'23) made extensive investigations at definite pH concentrations and found that, with the same oxygen content, different pH concentrations have definite effects on the time of survival of fishes. He also found that each species studied tolerated a rather wide range of pH with a fairly definite optimum for each species.

8-9

Ibid., pp.78-80. (5 through 9 were taken from Gutsell's thorough review of the literature dealing with the importance of pH and carbon dioxide for aquatic animals.)

10

Shelford, Victor E. "The Determination of Hydrogen Ion Concentration in Connection with Fresh-water Biological Studies," State of Illinois Department of Registration and Education, Division of Natural History Survey, Bulletin, Vol.XIV, Art.9, Feb., 1923.

METHOD AND PLAN OF STUDY

Preparation of Water

River water was used in all of the experiments. It tested approximately 8 p.p.m. dissolved oxygen, 5 p.p.m. free carbon dioxide, 65 p.p.m. fixed carbon dioxide, and a pH of 7.0 at room temperature. The oxygen was removed by heating the water in large open buckets in an autoclave at 240°F and 20 pounds pressure. At twenty minute intervals the pressure was released and the gases expelled by the violent boiling of the water. During the processing of the water, four-liter bottles in which the deoxygenated water was to be stored were set at the base of the autoclave and heat tempered. At the end of the heating period of two hours the pressure was again suddenly released and the boiling water siphoned from the bottom of the buckets to the bottom of the bottles. In this way the air in the bottles was floated out without becoming mixed with the incoming water. The bottles were immediately stoppered, sealed, and set aside to cool to laboratory temperature which varied between 26° and 29°C.

Water Analysis

Oxygen. A 250 cc glass-stoppered bottle was filled with the deoxygenated water immediately after unstopping a storage bottle and the dissolved oxygen content determined by the Modified Winkler method (11¹¹ '25).

Carbon dioxide. The determinations of carbon dioxide were made by the Seyler method. A 100 cc. sample of the water was analyzed for free carbon dioxide by adding phenolphthalein and titrating with N/44 sodium carbonate or N/44 hydrochloric acid solution depending upon whether the

¹¹

Standard Methods of Water Analysis, 1925.

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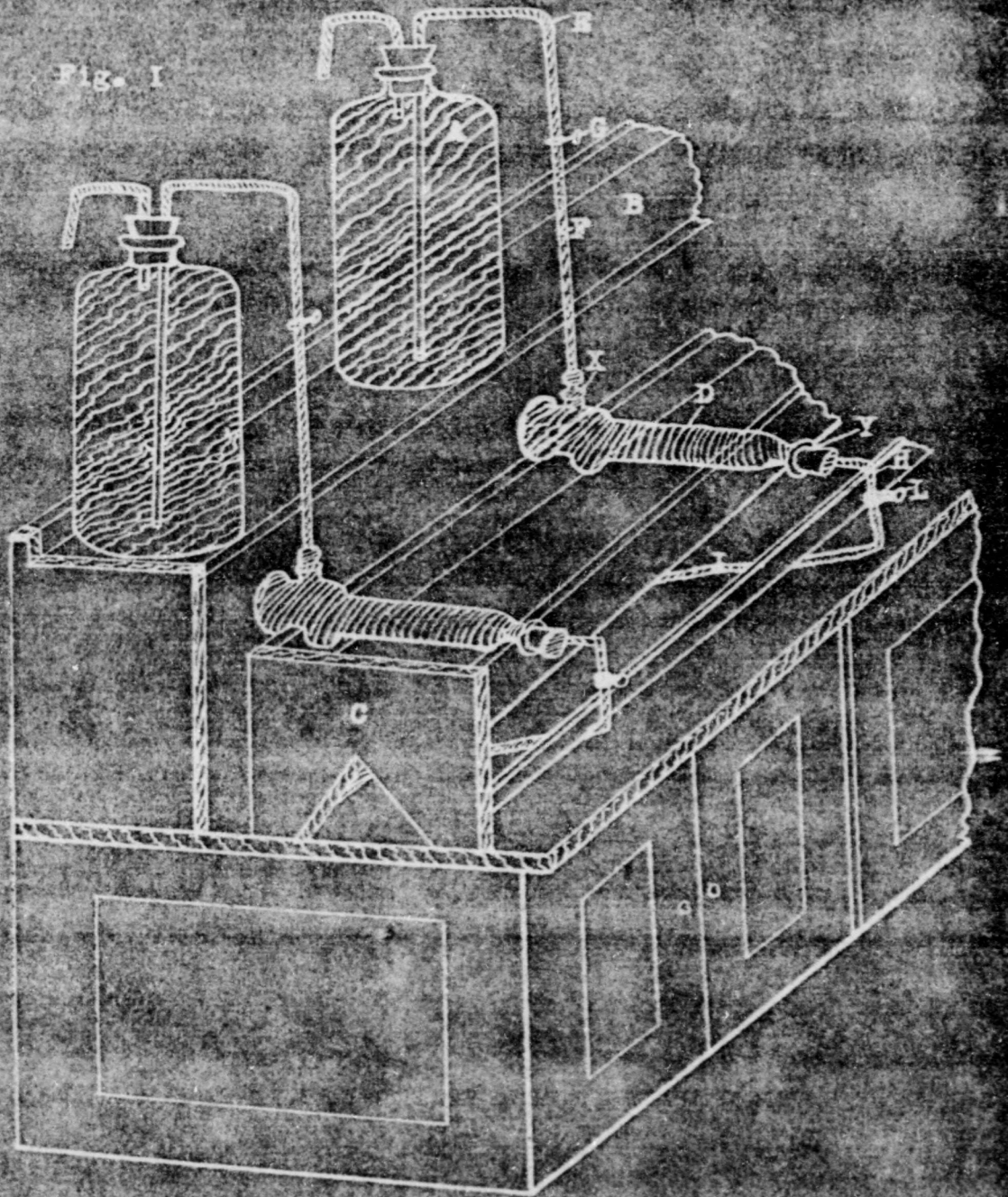
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APPARATUS FOR DETERMINING THE LETHAL POINT OF OXYGEN
AND CARBON DIOXIDE FOR FISHES

Fig. I



- A-- Reservoir Bottle
- B-- Shelf
- C-- Wooden Rack
- D-- Respiratory Chamber
(soda-lime tower)
- E-- Glass-tube with U-bend
- F-- Glass-tube
- G-- Pinch-cock

- H-- Right-angle Glass-bend
- I-- Long glass tube with
Right-angle bend
- L-- Screw Clamp
- X-- Opening at base of
Reservoir
- Y-- Opening at front of
Reservoir

water was acid or alkaline. A second sample of the water was analyzed for fixed carbon dioxide by adding methyl orange and titrating with ¹²N/44 - hydrochloric acid.

Hydrogen ion concentration. The pH was determined by the use of the Hellige Colcrimetric Apparatus.

The temperature of the water in each storage bottle was recorded at the beginning of each experiment.

Apparatus

The apparatus shown in the accompanying diagram was designed to hold and supply a medium in which the dissolved oxygen and carbon dioxide content in which fishes were to be tested could be controlled.

The reservoir bottles (A) were placed on a shelf elevated one and one-half feet above the laboratory desk (B). A wooden rack (C), constructed to hold eight soda-lime towers (D) which served as respiration chambers, was mounted on the laboratory desk. These towers were held horizontally with the larger opening of each tube forward and the smaller opening, which was at the base, directed upward. A glass-tube (E) with a U-bend at its upper end passed through the two-hole stopper in the neck of the bottle (A) and extended almost to the bottom of the reservoir. A short piece of rubber-tubing connected the short end of the U-tube (E) with a straight piece of glass-tubing (F), the lower end of which was held in the smaller opening of the respiration chamber by a tightly fitting one-hole stopper. The level of the respiration chamber was below the level of the base of the reservoir so that the water siphoned through the respiration chamber. The forward end of the respiration chamber was filled with a one-hole stopper in which was inserted a right-angle glass bend (H) which was

¹²

For further information of this procedure and the calculation of the carbon dioxide amounts see Birge, Edward A., and Juday, Chancy. "The Inland Lakes of Wisconsin," Wisconsin Geological and Natural History Survey, Bulletin XXII, Scientific Series No. 7, 1911, pp. 21-25.

connected by a short piece of rubber tubing to a second glass tube leading to the drain trough. A screw-clamp (L) was placed on the rubber tubing to regulate the rate of flow of the water through the respiration chamber. Another pinch-cock (G) facilitated the changing of reservoir bottles.

Procedure

A reservoir bottle was unstopped, the samples drawn for the water analysis, and the bottle set in place on the shelf. The two-hole rubber stopper with the glass tube (E) was inserted in the bottle, the pinch-cock (G) removed, and the water from the reservoir allowed to fill the respiration chamber. The rubber stopper (K) was loosened and the base of the chamber (D) raised so that all the air floated out at (X). The chamber was then flushed out in order to remove any aerated water and the specimen then introduced at either (X) or (Y) depending upon the size of the individual. The stopper was reinserted and the rate of flow of the water was regulated by screw-clamp (L) to 500 cc. per hour. Each specimen was kept in the respiration chamber for a period of three and one-half hours or more unless it died before this period of time had elapsed. A new specimen was used and the experiment repeated with increased oxygen content until a concentration was found that sustained the life of that species for a period of three and one-half hours. The lethal point was determined within 0.1 to 1 p.p.m. The dissolved oxygen content of the reservoir bottles was increased to the desired concentration by unstopping the bottles and shaking the contents exposed to the air.

Observations were made of the effects of free carbon dioxide tensions in the case of certain species of fishes. The free carbon dioxide content was increased to the desired concentration by bubbling carbon dioxide into the reservoir bottle.

TABLE 1. The Low Lethal Point For Certain Species of Fishes

Scientific Name	Common Name	Length of Specimen cm.	Temperature degrees C.	Lethal Point		
				Dissolved O ₂ conc.p.p.m.	Free CO ₂ p.p.m.	pH
1. <i>Ameiurus natalis</i>	Yellow Cat	7.5	28	1.04	0	7.2
2. <i>Cottus bairdii</i>	Miller's Thumb	7	26	0.942 to 1.57	3	7.4
3. <i>Clariicola</i> sp.?	Darter	7-8	26	1.199 to 1.285	3	7.4
4. <i>Notemigoneus crysoleucas</i>	Golden Shiner or Silverside	7-8	23	1.3 to 2.063	0	7.4
5. <i>Huro salmoides</i>	Large-mouthed Black Bass	7-9	26	1.673 to 1.799	0	7.5
6. <i>Apomctis cyanellus</i>	Green Sunfish	7-9	26	1.862	0	7.4
7. <i>Helioperca incisor</i>	Bluegill	9	27	1.315 to 2.672	0	7.2
8. <i>Chaenobryttus gulosus</i>	Warmouth Bass	7-8	27	2.055	0 to 1	7.4
9. <i>Notropis ardens lythrus</i>	Silverling	7-8	26	2.1415	0 to 3	7.2

RESULTS AND DISCUSSION

The nine species studied were *Ameiurus natalis*, *Cottus bairdii*, *Clariicola* sp.?, *Notemigoneus crysoleucas*, *Huro salmoides*, *Apomotis cyanellus*, *Helioperca incisor*, *Chaenobryttus gulosus*, and *Notropis ardens lythrurus*. Table 1 shows that there was but little variation in the length of specimens, in free carbon dioxide concentration, in temperature, or in pH range. The variations in these factors are believed to be too small to influence the lethal point for oxygen. Considerable variation in the concentration of dissolved oxygen, however, was found at the lethal point for the different species studied. A minimum of five and a maximum of fifteen specimens of the different species were used in determining the lethal point. Specimens were first placed in water of low dissolved oxygen content. If these specimens died, new specimens were then tested in water whose oxygen content was slightly higher. This procedure was repeated until water whose oxygen content would sustain the life of that species was found.

Ameiurus natalis (Yellow Cat). *Ameiurus natalis* lived for four hours in the respiration chamber in a dissolved oxygen concentration of 1.04 p.p.m. This specimen became dyspnoeic, excited, and distressed during the period of exposure but apparently recovered on being returned to the aquarium. On being subjected to a dissolved oxygen concentration of 1.27 p.p.m. the following day it succumbed within two and one-half hours. Due to the fact that other specimens of this species survived in this latter concentration, it was concluded that this specimen had been weakened by its treatment on the previous day. It was also concluded that *Ameiurus natalis* is able to live for a time in water containing a dissolved oxygen concentration as low as 1.04 p.p.m. at 26°C; that this concentration will eventually prove lethal; and that concentrations below 1.04 p.p.m. will not sustain life in this species longer than three hours.

No carbon dioxide tension determinations were made in the case of *Ameiurus* because Shelford and Allee (¹³'13) had previously found that this genus is narcotized in water containing 320 p.p.m. carbon dioxide and 1.43 p.p.m. dissolved oxygen.

Cottus bairdii (Miller's Thumb). *Cottus bairdii* lived in water of dissolved oxygen concentrations as low as 1.57 p.p.m. It died in water of 0.942 p.p.m. in 1 hour 40 minutes, while in concentrations of 0.642 and 0.856 p.p.m. the average length of life was from 10 to 15 minutes.

Of the nine species investigated *Cottus bairdii* seems to rank next to *Ameiurus natalis* in ability to survive water of low dissolved oxygen content. It is found lying quietly on the bottom of streams in the same habitat with *Clariicola* (one of the darters). In chambers of low oxygen concentration it shows little activity except for an occasional plunge forward, but at death it gives a sudden leap.

Clariicola sp.? (Darter). Specimens of this darter lived in the respiratory chamber in water containing a dissolved oxygen content of 1.285 p.p.m. Specimens died in less than one hour thirty minutes in water containing 1.199 p.p.m. dissolved oxygen.

Clariicola is a rather hardy darter which lives on the bottom of slowly moving streams. It has a comparatively small head and small gill surface. It seems to be as tolerant of low oxygen concentrations as *Cottus bairdii*. The fact that it is able to survive such low oxygen concentrations will doubtless be surprising to most people who are familiar with the darters. (The species is probably *whiplii*, but, since there is some doubt about this being true, it was thought best to omit the species name for this form.)

Notemigoneus crysoleucas (Golden Shiner or Silverside Minnow). Specimens of Notemigoneus crysoleucas easily tolerated water of dissolved oxygen content as low as 2.083 p.p.m., but died in less than three hours in water of 1.3 p.p.m.

This species survived increased carbon dioxide tensions up to and including 20 p.p.m. free carbon dioxide and will doubtless live in greater concentrations.

Notemigoneus crysoleucas is a rather hardy species which is found in many ponds of this region. It was, however, unable to live in water of low oxygen concentrations that supported Ameiurus natalis, Cottus bairdii, and Claricola.

Huro salmoides (Large-mouthed Black Bass). Huro salmoides survived exposure periods of three hours and thirty minutes in water as low as 1.799 p.p.m. dissolved oxygen but died within two hours in water at 1.673.

When the free carbon dioxide tension was raised to 100 p.p.m. with the dissolved oxygen remaining at 2.427 p.p.m. and the pH changed to 6.0, the specimen showed signs of distress and excitement but finally became quiet and lay on its side during the remainder of the experiment. It revived after it had been returned to the aquarium.

Huro salmoides is as hardy as Notemigoneus crysoleucas in regard to its ability to live in water of low dissolved oxygen content.

Apomotis cyanellus (Green Sunfish). Water of 2.328 p.p.m. dissolved oxygen content easily sustained this species while a concentration of 1.862 p.p.m. caused the specimens to lie on their sides and remain in that position for the remainder of the test period. The latter concentration would probably prove lethal. This sunfish died within two hours in a dissolved oxygen concentration of 1.354 p.p.m.

The free carbon dioxide tension was increased to 50 p.p.m. while the

oxygen was kept at 2.328 p.p.m. and the pH changed to 6.2. The specimen was greatly distressed but eventually became quiet for the remainder of the test period. The sunfish recovered in the aquarium.

Apomotis was caught in the same ponds with croppies, but the former easily existed in minnow buckets under conditions which proved fatal to the latter. From all observations these sunfish are almost as tolerant of low dissolved oxygen concentrations as *Huro salmoides*.

Helioperca incisor (Bluegill). *Helioperca incisor* proved less able to withstand low oxygen tensions than any of the species discussed above. It was able to live at 2.672 p.p.m. with ease, but it died in concentrations of 1.315 p.p.m. in less than one hour.

Increased free carbon dioxide tensions to 30 p.p.m. with the oxygen content remaining at 2.7 p.p.m. and the pH changed to 6.8 increased the respiration only to the hypernoic condition.

Chaenobryttus gulosus (Warmouth Bass). This species was not excited in concentrations above 2.055 p.p.m. dissolved oxygen. However, in this concentration it became distressed, darted about the tube, and finally came to lie on its side where it spent most of its time during the remainder of the test period. In an oxygen concentration of 1.104 p.p.m. it died within one hour.

The free carbon dioxide tension was raised to 75 p.p.m. while the dissolved oxygen was held at 3.122 p.p.m. and the pH changed to 6.0. The bass was barely able to survive the exposure periods in this concentration and remained on its side most of the time.

Notropis ardens lythrurus (Silverling). *Notropis ardens lythrurus* was barely able to survive the three hour and one-half periods of exposure in water containing 2.1415 p.p.m. dissolved oxygen. Any water of lower oxygen concentration than this will probably kill *Notropis*. Specimens died in water of dissolved oxygen content of 0.942 p.p.m. within 20 minutes.

SUMMARY

This investigation was undertaken for the purpose of determining the lethal point due to low concentrations of dissolved oxygen for certain species of fishes and for the observation of respiration changes brought about by increased free carbon dioxide tensions. A review of the literature shows that, while investigations of the effects of low dissolved oxygen concentrations on fishes have been made and the conclusions reached that the lethal point for any species is between zero and two parts per million, these studies were not carried on under carefully controlled laboratory conditions.

Specimens were placed in controlled individual respiration chambers through which water of various dissolved oxygen concentrations flowed at the rate of 500 cc per hour.

Results given in Table 1 show at least three levels of tolerance of fishes for dissolved oxygen concentration. No specimens of any species were able to live the duration of the test period in water at room temperature whose dissolved oxygen content was below 1 p.p.m. *Ameiurus natalis* and *Clariicola* sp.? belong to the highest level of tolerance which ranges from 1 to 1.3 p.p.m. *Cottus bairdii* lived for 1 hour and forty minutes in water of 0.942 p.p.m. dissolved oxygen which indicates that this species can, in all probability, survive the three and one-half hour test period in a dissolved oxygen concentration of 1.3. *Notemigoneus crysoleucas*, *Huro salmoides*, and *Apomotis cyanellus* belong to the second level which ranges from 1.3 to 2 p.p.m. *Helioperca incisor*, *Chaenobryttus gulosus*, and *Notropis ardens lythrurus* belong to the third or lowest level of tolerance which ranges from 2 to 2.3 p.p.m.

The data dealing with increased free carbon dioxide tensions is so limited that it is not offered as conclusive information even though it is believed to be correct. The results, however, are open to consideration. It was found that in waters ranging from zero to 10 p.p.m. free carbon dioxide there was no alteration of the eupnoic condition. *Notemigoneus crysoleucas* became hypernoic in water of 20 p.p.m. while 30 p.p.m. had similar effects on *Helioperca incisor*. Definite conditions of dyspnea existed, however, when *Euro salmoides* was placed in water of 100 p.p.m. at 2.427 p.p.m. dissolved oxygen concentration; when *Chaenobryttus gulosus* was placed in 75 p.p.m. free carbon dioxide at 3.122 p.p.m. dissolved oxygen; and when *Apomotis cyanellus* was placed in 50 p.p.m. free carbon dioxide at 2.328 p.p.m. dissolved oxygen. In the latter cases the specimens became excited and the respiration rate increased at once as soon as the fishes were placed in the chambers. This hypernoic condition was followed by a slowing down of the respiration rate and the loss of equilibrium. The fishes lay on their sides and respired spasmodically the remainder of the experiments but recovered after they were returned to the aquarium. The concentrations of free carbon dioxide were not increased to the extent that apnea resulted, but in the three cases of dyspnea it appeared that any increase in free carbon dioxide would prove lethal to the species involved.

ACKNOWLEDGMENTS

The writer wishes to acknowledge the two men who so willingly rendered their valuable services in the pursuit and completion of this investigation:

Dr. M.C.Ford, Director of Ogden Department of Science, directed the writing of the thesis.

Dr.L.Y.Lancaster, Professor of Zoology, directed the investigation and aided in the collection of specimens.

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