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## Hydrogen-ion Preferences and Tolerances of Certain Freshwater Fishes

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The main emphasis of this study was to determine whether fishes distinguish between or exhibit a preference for different pH values. Tolerance limits of certain fishes to various pH values were also tested. Powers (1932) states that within wide limits, the pH of water bathing the gills of aquatic animals has very little effect upon the organism. No specific range was mentioned. Powers' statement indicates that workers have experimented with the physiological tolerance of fishes to various pH values, but little information was found which might indicate pH preferences among fishes.

*Materials and Methods*—Specimens were placed within a preferential chamber and simultaneously confronted with aqueous media of different pH values. During the experimental period, the fish were free to position themselves to the media in any manner preferred.

The preferential chamber, described by Jones (1964), consisted of a cylindrical glass tube, 24 inches long and 2 inches internal diameter (Fig-

ure 1). The ends of the chamber were equipped with inflow glass tubes connected to two 5-gal aspirators. The inflow rate of a solution entering a particular end of the chamber could be slowed or reversed respectively by rearranging the position of screw clamps situated in the tubing complex. The water mixture was removed from the chamber by ventrally placed glass tubes, half way between the ends of the chamber. This outflow arrangement allowed a sharp gradient to exist between the two test solutions. The gradient was checked visually by injecting a red dye into one side of the chamber. The demarcation between the red and clear water remained sharp. The gradient was also checked chemically by removing samples from the chamber through the air ports. The pH value in each side of the chamber varied from its source by approximately 0.2 unit on the pH scale.

The two aspirators were filled with water taken from the same source. They differed in that one contained a small amount of 36% HCl. This acid was selected as the agent to increase the pH because it has a high level of dissociation and because chloride anions would be less likely to cause adverse effects on fish than other organic or inorganic anions.

Hydrogen-ion concentrations in the nonmodified water ranged from pH 7.6 to 8.2. The water in both aspirators was agitated before entering into the preference chamber. Agitation aided to eliminate CO<sub>2</sub> in the water and to equalize the available oxygen supply in both aspirators. The Beckman Zeromatic II pH meter was used for pH determinations.

Positions of the fish in the chamber were recorded at 1-min intervals on a chart which schematically represents the chamber (Figure 2). Any vertical line on the chart represents the fish in a quiescent position in the corresponding area in the tube where the behavior occurred, and the accompanying minutes on the ordinate of the chart indicates the time spent during such behavior. Likewise, horizontal lines represent the fish in a moving state. Sixteen tests, each lasting 32-40 min, were conducted. During this time, the contents of the aspirators were randomly switched from one end of the chamber to the other. By maneuvering the water supplies in this manner, experimental bias was kept to a minimum.

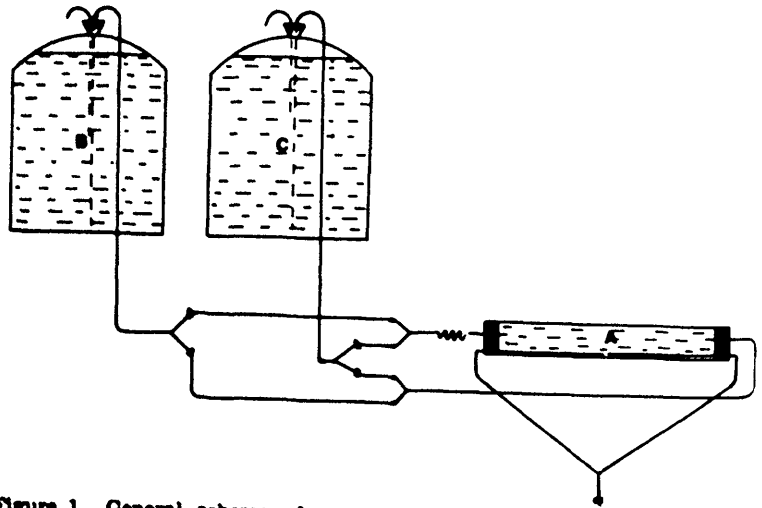


Figure 1. General scheme of apparatus for observing the reactions of fish to pH values. Fish were confined within the chamber, A; aspirators B and C supplied water to the chamber.

TABLE I. OBSERVATIONS ON THE REACTIONS OF CERTAIN FISHES TO VARIOUS pH VALUES.

Group	Species	Observations at one minute intervals in:			Sum of low pH	Sum of high pH
		Low pH	High pH			
I (pH 6.0 ± 0.2 vs. 8.0 ± 0.2)	Cyprinus carpio	19	15			
	Fundulus kansae*	66	3			
	Fundulus kansae	35	8			
	Chrosomus erythrogaster*	39	30	159	56	
II (pH 5.0 ± 0.2 vs. 8.0 ± 0.2)	Etheostoma spectabile	6	30			
	Notropis lutrensis	16	16			
	Cyprinus carpio	3	25	35	71	
III (pH 3.0 ± 0.2 vs. 8.0 ± 0.2)	Fundulus kansae*	6	65			
	Fundulus kansae	8	23			
	Cyprinus carpio	9	31			
	Lepomis macrochirus	5	31			
	Notropis lutrensis	38	37			
	Gambusia affinis**	16	16			
	Ictalurus natalis**	16	16			
	Lepisosteus sp.*	33	34	131	253	
				225	380	

\*two individuals/test

\*\*data altered, no preference

For the pH tolerance tests, aquaria were set up containing solutions with the same pH values as the water used in the preference chamber. These were stocked with several species of fishes and observed for 24 hrs. The pH values of the test solutions were not maintained because the continuous nitrogenous waste produced by the fish buffered the water.

Species tested were: *Etheostoma spectabile*, *Lepomis macrochirus*, *Micropterus salmoides*, *Fundulus kansae*, *Gambusia affinis*, *Ictalurus*

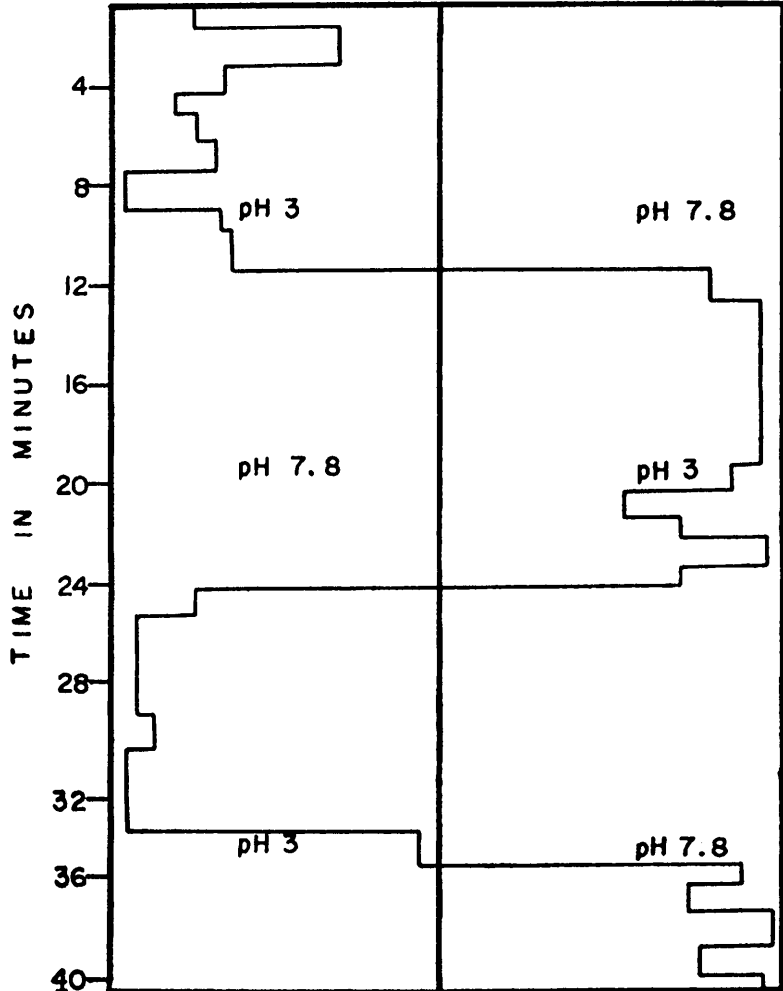


Figure 2. Reactions of a carp to waters of different pH values. Time is recorded in minutes on the ordinate and the figures indicate the pH values tested. Thus, the experiment was started with water of pH 3 flowing in on the left side; after 19 minutes water of pH 7.8 was introduced from the left side.

*natalis*, *Notropis boops*, *Notropis lutensis*, *Campostoma anomalum*, *Chrosomus erythrogaster*, *Cyprinus carpio*, and *Lepisosteus* sp.

**Results and Discussion** — Reactions of fishes to various pH values are presented in Table I and Figure 2. There were 380 observations (62.5%) made of fishes in the end of the chamber possessing alkaline waters compared to 225 observations (37.5%) in acidic waters. For a more detailed account, the preference tests were divided into three groups.

Test species in Group I exhibited a slight preference for the acidic water ( $\text{pH } 6.0 \pm 0.2$ ), although the preference was significant only for the plains killifish, *F. kansae*. Species of Group II, which were subjected to hydrogen ions 10 times more concentrated than Group I, reacted negatively to the acidic water. The red shiner, *N. lutensis*, however, exhibited no observable preference. Species of Group III presented with pH values of  $3.0 \pm 0.2$  vs  $8.0 \pm 0.2$ , 1000 times more acidic than concentrations tested in Group I, generally reacted negatively to the acid condition. Four of the species in Group III did not exhibit observable preferences.

TABLE II. TOLERANCE TESTS OF CERTAIN FISHES TO DIFFERENT pH VALUES.

pH 3.1	pH 5.5
<i>Gambusia affinis</i> *	<i>Gambusia affinis</i>
<i>Fundulus kansae</i> *	<i>Fundulus kansae</i>
<i>Cyprinus carpio</i> *	<i>Cyprinus carpio</i>
<i>Lepomis macrochirus</i> *	<i>Lepomis macrochirus</i>
	<i>Campostoma anomalum</i>
	<i>Etheostoma spectabile</i>
<i>Ictalurus natalis</i>	<i>Ictalurus natalis</i>
<i>Lepisosteus osseus</i> *	
	<i>Chrosomus erythrogaster</i>
<i>Notropis lutensis</i> *	
pH 6.4	pH 8.2
<i>Gambusia affinis</i>	<i>Gambusia affinis</i>
<i>Fundulus kansae</i>	<i>Fundulus kansae</i>
<i>Cyprinus carpio</i>	<i>Cyprinus carpio</i>
<i>Lepomis macrochirus</i>	<i>Lepomis macrochirus</i>
<i>Campostoma anomalum</i>	<i>Campostoma anomalum</i>
<i>Etheostoma spectabile</i>	<i>Etheostoma spectabile</i>
<i>Ictalurus natalis</i>	
	<i>Lepisosteus osseus</i>
<i>Chrosomus erythrogaster</i>	<i>Chrosomus erythrogaster</i>

\*died

The reactions and behavior of the carp, *C. carpio*, to pH values of 3.0 vs 7.8 are depicted in Figure 2. At the beginning of the experiment, acid water was introduced into the end of the chamber the fish occupied. The fish remained in the acidic water for 11 min before it crossed the boundary into alkaline water. The fish appeared calm in the alkaline water and remained in the right side of the chamber until acidic water was introduced. The fish became restless, appearing to detect the acidic condition of the water and, while randomly swimming, crossed the boundary back into alkaline water.

The two test solutions were randomly exchanged between ends of the chamber throughout the experiment; however, the carp consistently selected the alkaline water.

Results of the pH tolerance tests are shown in Table II. During a 24-hr period, death occurred only in fishes subjected to extremely acidic conditions (pH 3.1). This concentration was lethal to all species tested except the yellow bullhead, *I. natalis*. Death of these fishes might be comparable to increased CO<sub>2</sub> tensions of the blood, associated with lower pH of the blood (Lagler et al., 1962). This condition could possibly result in asphyxiation. Another potential cause of death might have been the disruption of the buffering system, with a possible breakdown of enzyme complexes.

Although preliminary, these data indicate that certain fishes exhibit a preference in regard to the pH of their environments. The rapidity with which they reacted to pH changes seems to be related to their sensitivity, as determined by tolerance tests, and also perhaps to the degree that the pH varied from the water in which they were normally found.

#### LITERATURE CITED

- Jones, J. F. R. 1964. *Fish and River Pollution*. Butterworth & Co., Ltd. London. 203 p.
- Lagler, K. F., J. E. Bardach, and R. R. Miller. 1962. *Ichthyology*. John Wiley and Sons, Inc., New York. 545 p.
- Powers, Edwin B. 1932. The relation of respiration of fishes to environment. *Ecol. Monogr.* 2:387-473.
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