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EFFECTS OF TEMPERATURE, LIGHT, FEEDING,
AND ACTIVITY ON THE RATE OF AERIAL
BREATHING OF GAR (LEPISOSTEUS).

The University of Oklahoma, Ph.D., 1963
Zoology

University Microfilms, Inc., Ann Arbor, Michigan

THE UNIVERSITY OF OKLAHOMA
GRADUATE COLLEGE

EFFECTS OF TEMPERATURE, LIGHT, FEEDING, AND ACTIVITY ON THE
RATE OF AERIAL BREATHING OF GAR (LEPISOSTEUS)

A DISSERTATION
SUBMITTED TO THE GRADUATE FACULTY
in partial fulfillment of the requirements for the
degree of
DOCTOR OF PHILOSOPHY

BY
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1963

EFFECTS OF TEMPERATURE, LIGHT, FEEDING, AND ACTIVITY ON THE
RATE OF AERIAL BREATHING OF GAR (LEPISOSTEUS)

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ACKNOWLEDGMENTS

I wish to express my deep appreciation and gratitude to Dr. Carl D. Riggs for stimulation and encouragement during this investigation and for providing invaluable aid in the writing of the manuscript; to Mr. W. Frank Wade, a graduate student in the Department of Zoology for indispensable aid on several occasions; to Dr. Arthur W. Ghent for help and advice with the statistical analysis of the data; to several students at the University of Oklahoma Biological Station for helping me collect the gars; to the University of Oklahoma Alumni Foundation for financial help in preparation of the dissertation; and finally to my wife Sudha, whose continual encouragement has meant so much to me.

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EFFECTS OF TEMPERATURE, LIGHT, FEEDING, AND ACTIVITY ON THE
RATE OF AERIAL BREATHING OF GAR (LEPISOSTEUS)

CHAPTER I

INTRODUCTION

Air breathing of fishes has aroused the curiosity of biologists for more than 100 years. In the New World, air-breathing fishes include Lepidosiren, Arapaima, Erythrinus, Amia, and the extant species of the genus Lepisosteus commonly called gars. Gars are found in the fresh waters of North America east of the Rocky Mountains, south to Guatemala, Cuba, and the Lake Nicaragua basin, and on the Pacific slope of middle America from southern Mexico to Honduras (Moore, 1957). Fossils of this genus have been reported from upper Cretaceous deposits to those of lower Miocene in Europe; in North America from middle Eocene rocks to Recent; and from Eocene deposits in Asia (Berg, 1947; Romer, 1945).

In order to ventilate the gas bladder or lung, gars approach the surface and thrust the elongated jaws and part

of the head out of the water. This habit, along with the primitive morphology of these fishes, stimulated the publication of such early papers as those of Poey (1856, 1858), Agassiz (1859), Wilder (1876, 1877a, 1877b), Agassiz (1879), Balfour and Parker (1881), Parker (1881), Mark (1890), and Morris (1886). The first biologists to describe this behavior in Lepisosteus were not certain of its purpose, although they suspected it to be concerned with air breathing (Poey, 1856, 1858; Agassiz, 1859; Wilder, 1876; Mark, 1890; Potter, 1926). The first good experimental evidence that the gars actually are air breathers was provided by Potter (1927), and C. D. Riggs and G. A. Moore (unpublished) recently have added even more conclusive evidence as well as a general description of the mechanism involved in pulmonary ventilation.

The relationship between the respiratory functions of gills and lungs (or respiratory gas bladders) in air-breathing fishes is poorly understood. Carter (1957) stated that the occurrence of air-breathing organs is accompanied by reduction of the gills. George and Dubale (1941) reported that the respiratory area of the gills in air-breathing fishes is on the average less than one half of that of the water-breathing fishes. This reduction in gill area has not

been determined for Lepisosteus.

The effects of both intrinsic and extrinsic factors upon aerial breathing rate of air-breathing fishes are poorly understood. Although much is known about the effects of internal and external environmental factors on the respiration of air-breathing tetrapods, it does not necessarily follow that these data are applicable in the same way to air breathing of the more primitive fishes.

Although the evidence that gars are obligatory air breathers, at least during the summer months, is quite conclusive, there is at present no explanation for the irregularity of aerial breathing during a 24-hour period. This irregularity has been observed many times, both in the laboratory and in the natural environment, during periods when there is no apparent appreciable change in conditions of the external environment. The chief purpose of this study is to measure the effects of four common extrinsic factors (temperature, light, feeding, and activity) on the rate of aerial breathing of Lepisosteus, in order to provide some explanation of the irregular pattern of aerial breathing and to provide additional evidence that gars are air breathers and that this activity is of vital physiological importance to them. This work is a part of a comprehensive study of the aerial breath-

ing of Lepisosteus and the bowfin, Amia calva, being conducted at the University of Oklahoma Biological Station and on the Norman campus under the direction of Drs. Carl D. Riggs and George A. Moore.

CHAPTER II

MATERIALS AND METHODS

Most of my research was done with the longnose gar, L. osseus. When individuals of this species were not available the spotted gar, L. oculatus, was used. Ten longnose and five spotted gar were available at the beginning of the research. Five of the former and one of the latter died during the preliminary stages of the work, and during the course of the critical experiments two additional longnose gar died of undetermined cause. Attempts were made to obtain additional fish of the proper size, but none could be located, chiefly due to the fact that so little is known of the habits of gars in winter. These gars were collected from Lake Texoma, an impoundment of the Red and Washita rivers, Oklahoma and Texas, during the summer of 1961 and were all yearlings of the 1961 year-class. The average total-lengths of the longnose and spotted gars were 382 mm and 280 mm respectively. During the time the experiments were

performed each gar was fed live minnows approximately every four days.

My work began in October, 1960 in a search for and the testing of adequate instruments for measurement of aerial breathing of gars, and ended with the feeding experiments in February, 1963. The main problem at the beginning of the research was to find a method of continuously recording the rate of aerial breathing during the varying conditions. This was essential for measurements in darkness, and highly desirable in light in order to eliminate the possibility of human error. Since gars inevitably create at least a slight disturbance of the surface of the water when they breathe air, the solution seemed simple and obvious, i.e., devise a method of recording even slight disturbances on the surface of the water without disturbing the fish. Since the gars were to be kept in aquaria throughout the experiments, this meant that only small surfaces were involved.

I first attempted to use an electrical kymograph with tambours and various kinds of floats of cork and balsa wood. This apparatus was not sufficiently sensitive and was discarded. Next, through the interest of a fellow graduate student, Mr. Ziad Shehadeh, I learned of the possibilities of the use of strain gauges in recording any source of strain

on bendable objects. Professor James O. Melton, of the Aeronautical Engineering Department discussed the uses of strain gauges with me and agreed to provide a strain gauge analyzer and an electronic recorder (oscillograph) and the necessary strain gauges for some preliminary experiments. He mounted these gauges toward one end of slender strips of stainless steel.

A ping pong ball was attached to one end of the metal strip to act as a float on the water surface; the other end was firmly fixed to the edge of the aquarium with a C-clamp. Whenever the gar took a breath the wave action it created disturbed the ping pong ball on the water surface and this disturbance was recorded by the oscillograph. This method proved to be quite successful but because the equipment had been heavily used and was not in good repair it did not work consistently. When this apparatus did work it worked well (Figure 1) but its inconsistency and the cost of purchasing the proper new equipment resulted in its being discarded.

The possibility of using photoelectric cells to record breathing was explored next. A Physiograph 3-channel recorder, which uses a photoelectric cell in certain transducers, was tested through the courtesy of the manufacturer, the E & M Instrument Company of Houston, Texas. The results

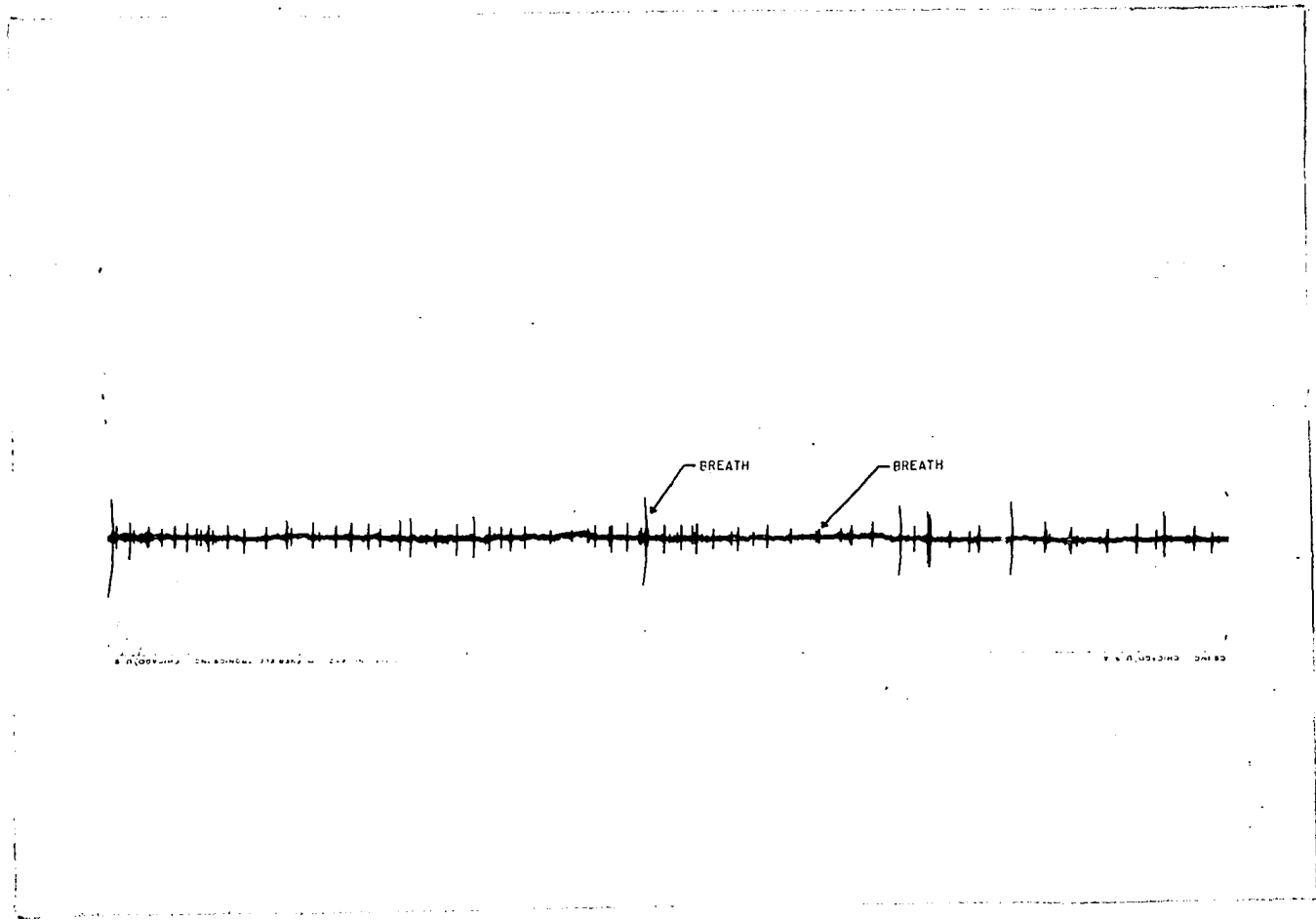


Figure 1. A typical recording of aerial breathing of a gar made by the strain-gauge apparatus.

were excellent. The Physiograph and the necessary accessory equipment was purchased from grant monies provided to Dr. Carl D. Riggs by the University of Oklahoma Research Institute and the University of Oklahoma Alumni Development Fund.

The Physiograph produces multi-channel infinite-length recordings with all events time-correlated on the graph paper. As I used it, a recording channel consisted of a transducer, an A.C.-coupler to filter out 60 cycles disturbance, an amplifier, and a pen recorder. Physiological events are converted into proportional electrical signals by the transducer. These signals are increased in intensity by the amplifier and used to energize the recording pen to produce a permanent graphic record. The transducer was activated by the movements of a ping pong ball suspended by a thin nylon thread from the hook on the shutter of the transducer. The thread was adjusted so that it was taut and allowed the ball to float on the water. Whenever the gar took a breath it disturbed the water surface and jiggled the ball; this opened and closed the shutter on the transducer, activating it, and the disturbance was recorded on the graph paper. A typical recording is shown in Figure 2. The time marker of the Physiograph was set to mark on the graph paper at one-minute intervals and thus the exact time of each breath

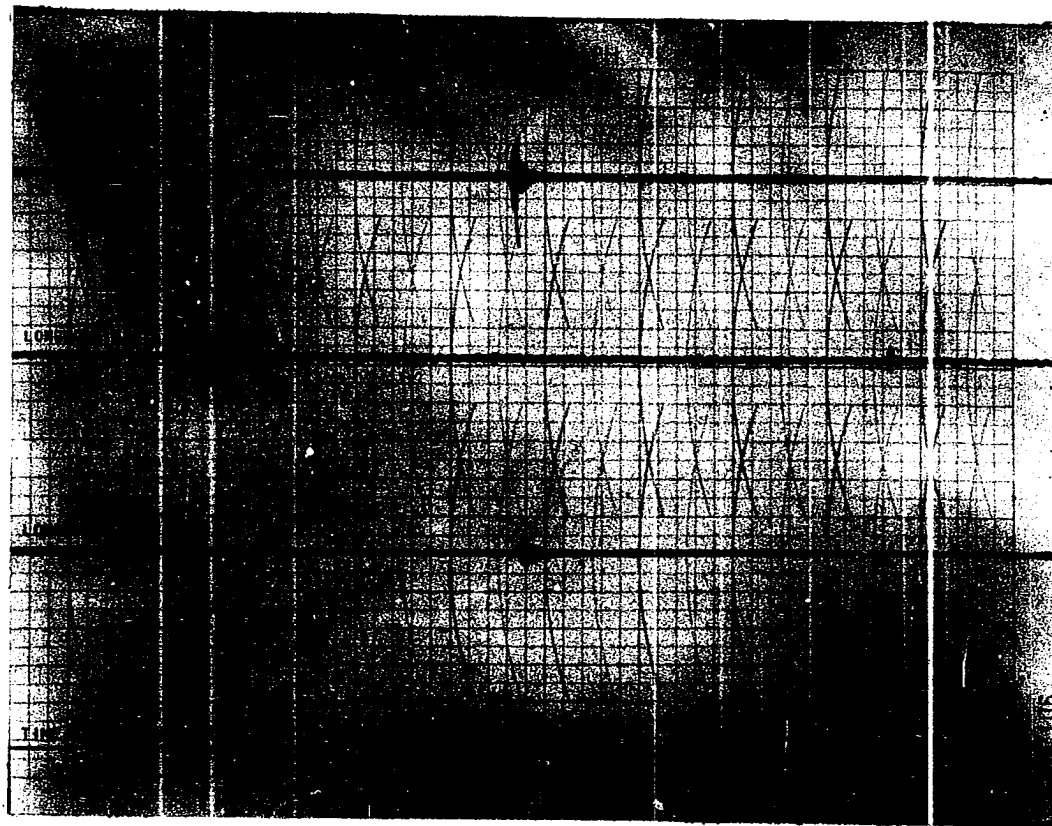


Figure 2. A typical recording of aerial breathing of gars made by the Physiograph.

and the intervals between breaths were easily calculated. The set up of the apparatus is shown in Figures 3 and 4.

The entire research was conducted in a controlled temperature room, approximately 10 x 7 x 8 feet, with thick, insulated, black walls. The room was lighted by two 48T12, fluorescent, 60-watt, cool white tubes each 46 inches long. The lights were plugged to an automatic timer device with which they could be turned off and on at any desired interval. The desired temperature in the room was maintained by a thermostatically controlled refrigeration unit with a temperature range of 30 to 70 F. The temperature of the water in the aquaria was raised above 70 F with 250-watt aquarium heaters and was regularly checked with floating thermometers, which floated in the aquaria throughout the experiments.

Three aquaria (one of 35 gallons and two of 50 gallons) each with one fish were used for the major portion of the research. Near the end of this research the 35-gallon aquarium was replaced by one with a 50-gallon capacity. Application of the paired t test showed that the rate of aerial respiration was not significantly affected by the different size aquaria (Appendix Table 1). The three channels of the Physiograph recorder limited the number of aquaria that could be used and thus the number of fish that could be tested at

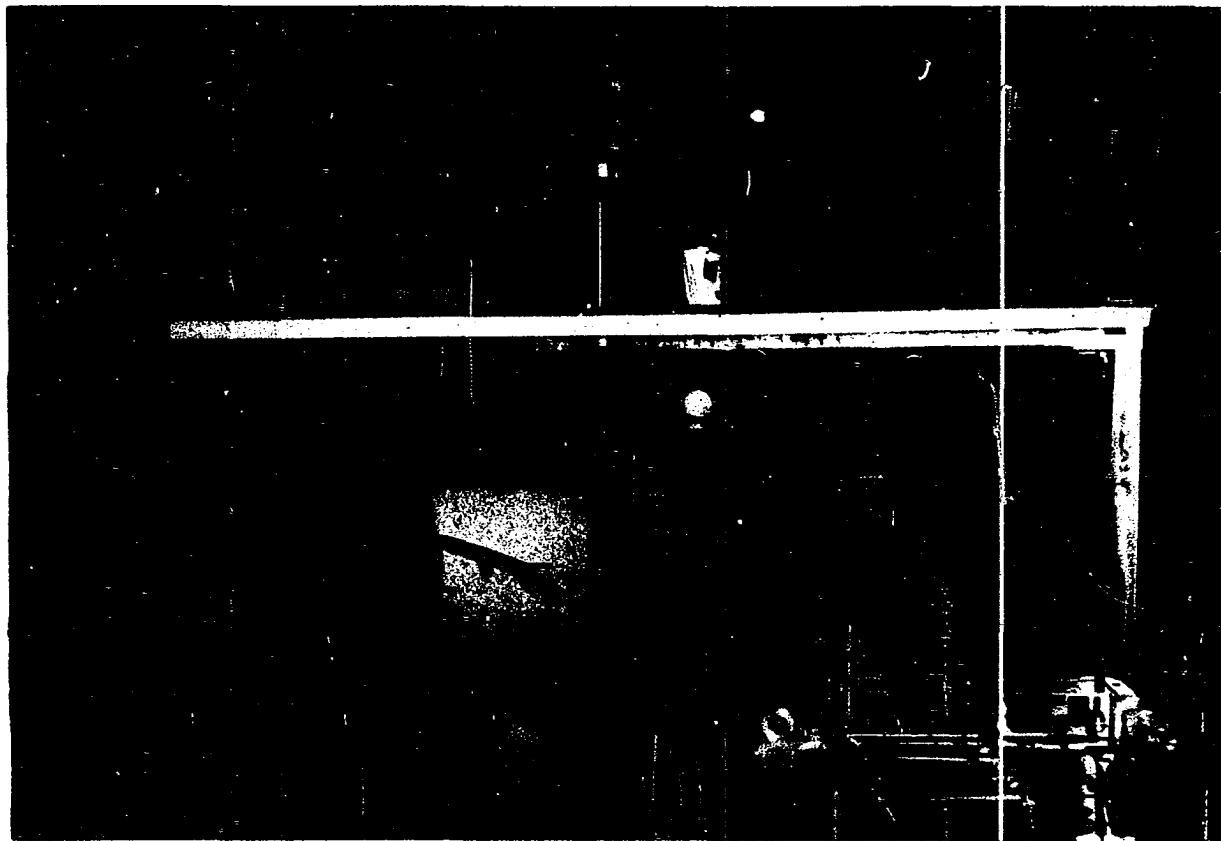


Figure 3. The experimental set up including the Physiograph and other accessories for recording the aerial breathing of gars.

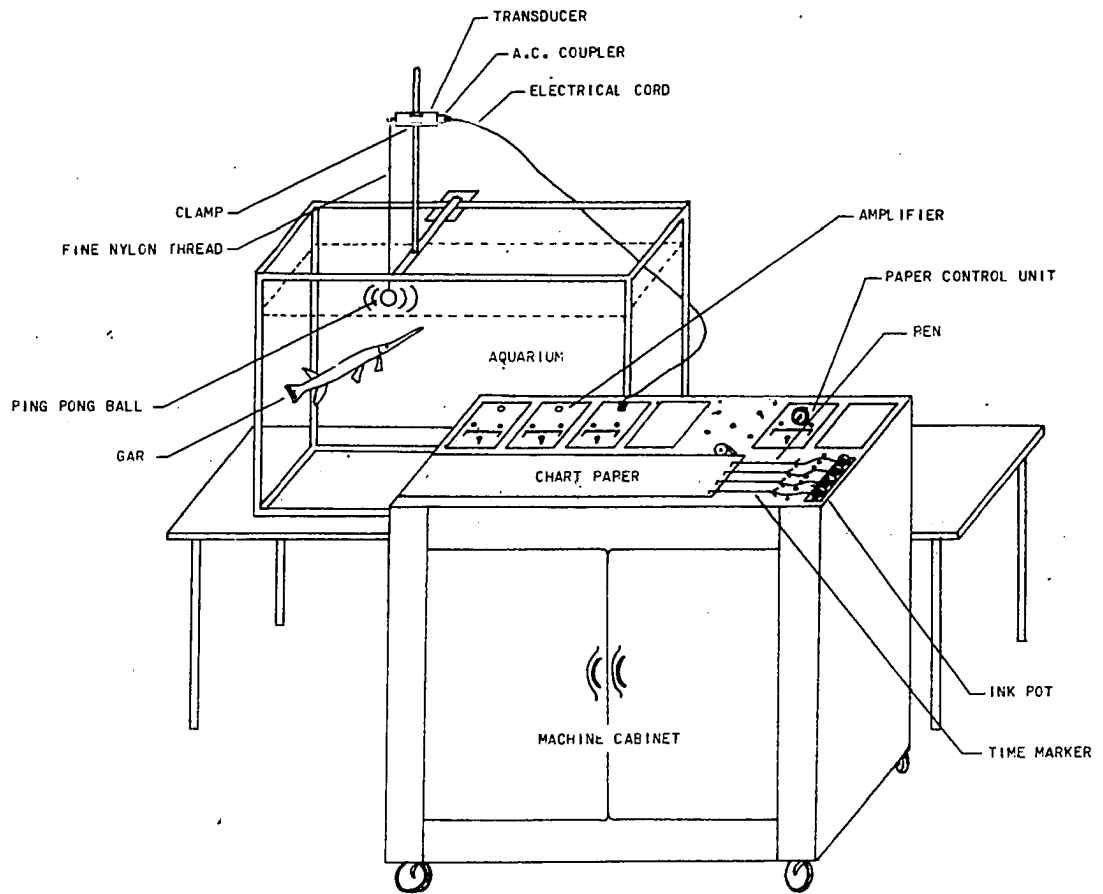


Figure 4. Diagrammatic sketch of the experimental set up including the Physiograph for recording the aerial breathing of gars.

any one time. This limitation, along with the fact that the temperature experiments were performed first, necessitating the use of the same three fish, if possible, throughout the long series (102 days), and the unexplained mortality of reserve fish, resulted in the use of a limited number of individuals throughout the work. The tops of the aquaria were covered with screen wire and polyethylene sheeting to prevent the disturbance of the water surface by air coming from the refrigeration unit fan. An opening (12 x 6 inches) was provided on one side of the cover for aeration. Except for the top covers the aquaria were not shielded on any other side. Laboratory tap water having the freezing point depression of 0.05 F was used in these aquaria.

CHAPTER III

EFFECT OF TEMPERATURE

Procedure

Both the longnose and spotted gars were used in the experiments designed to determine the effects of different temperatures on the rate of aerial respiration. Three longnose gars, designated as A, B, and C, were exposed to temperatures of 38, 42, 46, 50, 54, 60, 66, 72, 86, and 96 F for an average of four days (Table 1) at each temperature, and the breathing rates simultaneously recorded. Fish A died of an unknown cause during the experiment while the temperature was 66 F and, since no other longnose gar of the same approximate age and weight was available, was replaced by a spotted gar of the same year-class. Fish C died at 86 F and was not replaced because no other gar of the same age and weight was available. Temperatures to which the spotted gar was exposed were 50, 60, 72, 90, 100, and 104 F, also for an average of four days each (Table 1). Not knowing the extremes of

temperature which these fish could endure, 72 F was chosen arbitrarily as the starting point and the temperature was lowered stepwise by 4 to 6 F intervals until aerial respiration virtually ceased (38 F) and the fish depended upon aquatic respiration. The temperature was then allowed to return gradually to 72 F and the fish were acclimated for two weeks. It was then raised stepwise to the highest temperatures used (96 F for the longnose gar; 104 F for the spotted gar). Each gar died at these respective temperatures.

From 38 to 72 F the temperature of the water in the aquaria was the same as that of the room in which they were kept; above 72 F aquarium heaters were used to raise the water to the desired temperature. The room was lighted from 6:30 A.M. to 6:30 P.M. and dark from 6:30 P.M. to 6:30 A.M. throughout the temperature experiments. During the successive changes in temperatures the fish remained in the aquaria and a period of at least 24 hours was allowed for the fish to become acclimated to the new temperature after each temperature change. Most of the periods of acclimation were used for feeding the fish. Throughout the experiments visual observations were recorded to supplement the records of the Physiograph and to note whether there was any change in the general behavior of the fish.

Results

In general the rate of aerial respiration increased as the temperature increased (Table 1) for both the longnose gar (Figures 5, 6, 7) and the spotted gar (Figure 8). The increase was neither uniform nor consistent. The breathing rate for the longnose gar decreased in the temperature intervals of 66 to 72 F (fish B and C; Figures 6 and 7) and 86 to 96 F (fish B; Figure 6); that of the spotted gar decreased in the temperature interval of 72 to 90 F (Figure 8); above 72 F for the longnose gar and 90 F for the spotted gar the rate of aerial breathing increased again. It is possible that the decreases that occurred at the temperature intervals of 66 to 72 F and 72 to 90 F are artificial, since in each case the 72 F point occurred within the interval. I do not believe that these decreases are artificial because in the case of longnose gar it occurred below 72 F as the temperature was being decreased, and in the spotted gar it occurred above 72 F as the temperature was being increased; in the longnose gar B the decrease occurred between 86 and 96 F when the temperature was being increased. Such fluctuations with several other fishes have been reported in the literature and more work is needed to find explanation for these inconsistencies.

TABLE 1

Average breathing rates of longnose and spotted gars at different temperatures

	Longnose Gar A (Average Breaths/Hour)	Longnose Gar B (Average Breaths/Hour)	Longnose Gar C (Average Breaths/Hour)	Spotted Gar (Average Breaths/Hour)
38 F	0.029	0.006	0.053	
Standard Error	0.010	0.005	0.016	
Number of Trials*	14	14	14	
42 F	0.048	0.048	0.166	
Standard Error	0.010	0.015	0.036	
Number of Trials	12	12	12	
46 F	0.141	0.111	0.133	
Standard Error	0.031	0.041	0.086	
Number of Trials	10	9	10	
50 F		0.062	0.145	0.249
Standard Error		0.007	0.019	0.029
Number of Trials		12	12	14
54 F	0.991	0.431	0.618	
Standard Error	0.298	0.111	0.122	
Number of Trials	8	8	8	
60 F		2.327	0.673	2.356
Standard Error		0.121	0.048	0.334
Number of Trials		14	12	14

TABLE 1--Continued

	Longnose Gar A (Average Breaths/Hour)	Longnose Gar B (Average Breaths/Hour)	Longnose Gar C (Average Breaths/Hour)	Spotted Gar (Average Breaths/Hour)
66 F	4.128	6.277	3.785	
Standard Error	0.509	0.708	0.115	
Number of Trials	6	6	7	
72 F		5.124	3.430	5.798
Standard Error		0.626	0.779	0.526
Number of Trials		12	12	12
86 F		11.041	10.437	
Standard Error		0.609	0.687	
Number of Trials		8	8	
90 F				3.666
Standard Error				0.182
Number of Trials				7
96 F		7.416		
Standard Error		0.282		
Number of Trials		8		
100 F				9.135
Standard Error				1.278
Number of Trials				8
104 F				15.416
Standard Error				0.833
Number of Trials				2

*Trial = 12 hours.

At the lowest temperature tested (38 F) the aerial breathing rate for the longnose gar was measured in hundredths (fish A and C) or thousandths (fish B) of breaths/hour which is practically no aerial respiration. The rate increased as the temperature was raised to 50 F, but was still low (in hundredths or tenths breaths/hour). Above 50 F the rate of aerial breathing increased markedly. In Oklahoma, gars in their natural habitat are never seen to come to the surface to breathe in the winter. At low temperatures ranging from 38 to 50 F, they apparently remain in deeper water and do not surface for aerial breathing. Both the longnose and spotted gars died at 104 F which may be the lethal limit of temperature for the species.

At the lower (38 to 50 F) and higher (above 90 F) temperatures the experimental fishes ate very little (less than two minnows in four days) and usually remained practically motionless on the bottom of the aquaria except when they came to the surface to breathe. Their maximum activity and feeding (more than six minnows per day) occurred between 66 to 75 F. This seemed to be the optimum range of temperature for their normal activity and fish exposed to temperatures in this range were usually found near the surface in the night but as soon as the lights were turned on they went

toward the bottom of the aquaria. In the temperature ranges of 38 to 50 F and above 90 F, the gars seldom were near the surface (except, of course, for aerial respiration) but if they were seen near the surface at night they went to the bottom of the aquaria as soon as the light was turned on.

Discussion

The belief is prevalent among biologists that metabolism and rate of oxygen consumption of the cold-blooded vertebrates increase as the temperature of the environment increases, at least until sublethal temperatures are closely approached. In general, the results of this study seem to agree with this belief although the breathing rate did not consistently increase, but decreased during the temperature increase from 66 to 72 F (fish B and C), 86 to 96 F (fish B), and 72 to 90 F (spotted gar).

Numerous studies on the effect of temperature on aquatic respiration of fishes, measured in terms of oxygen consumption, activity, and general metabolism, are reported in the literature and show comparable results. Fry and Hart (1948), working with goldfish, found that the sustained swimming rate drops at temperatures from 30 to 38 C, probably because of a decrease in the metabolism available for

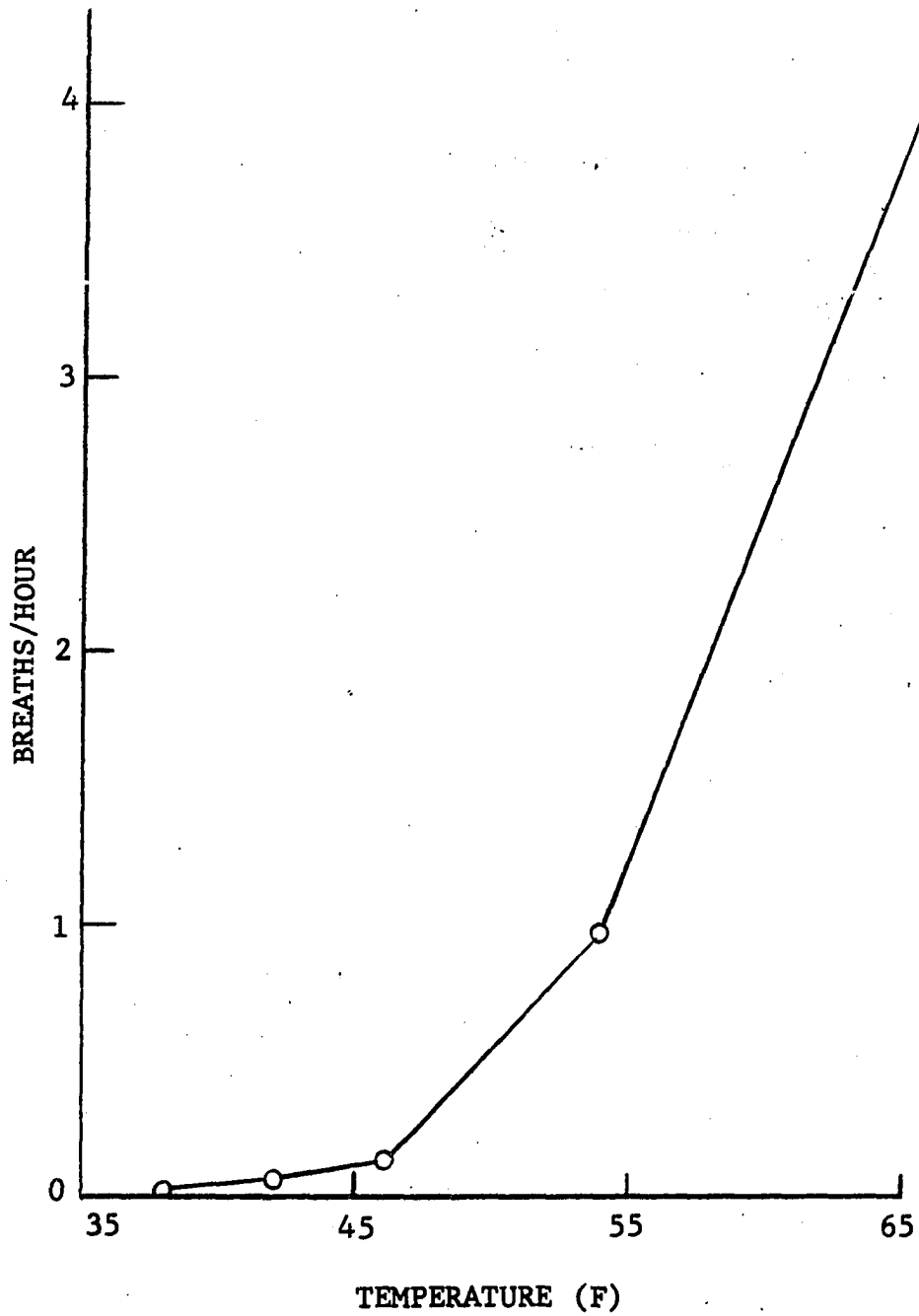


Figure 5. Rate of aerial breathing of longnose gar A at different temperatures.

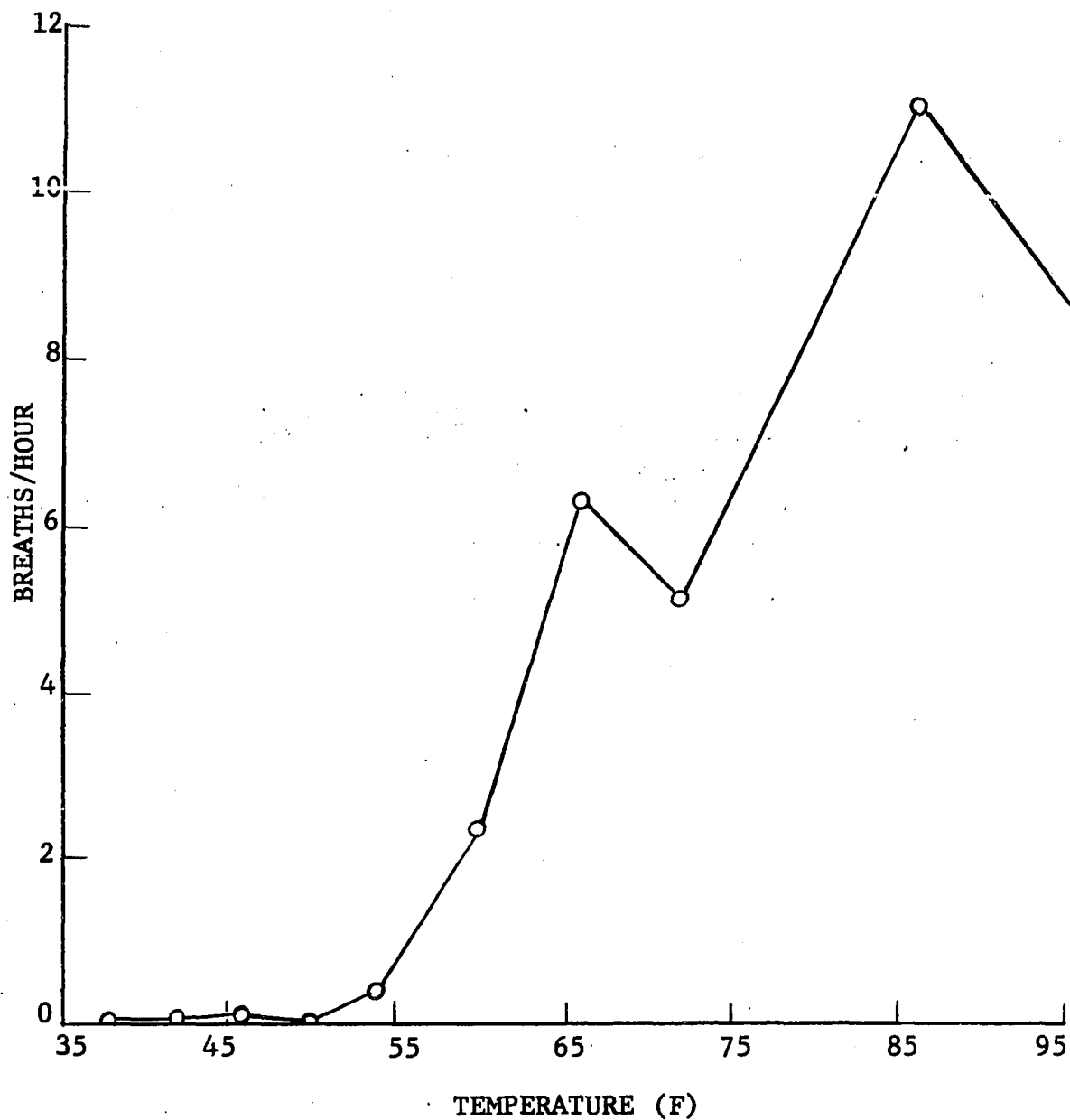


Figure 6. Rate of aerial breathing of longnose gar B at different temperatures.

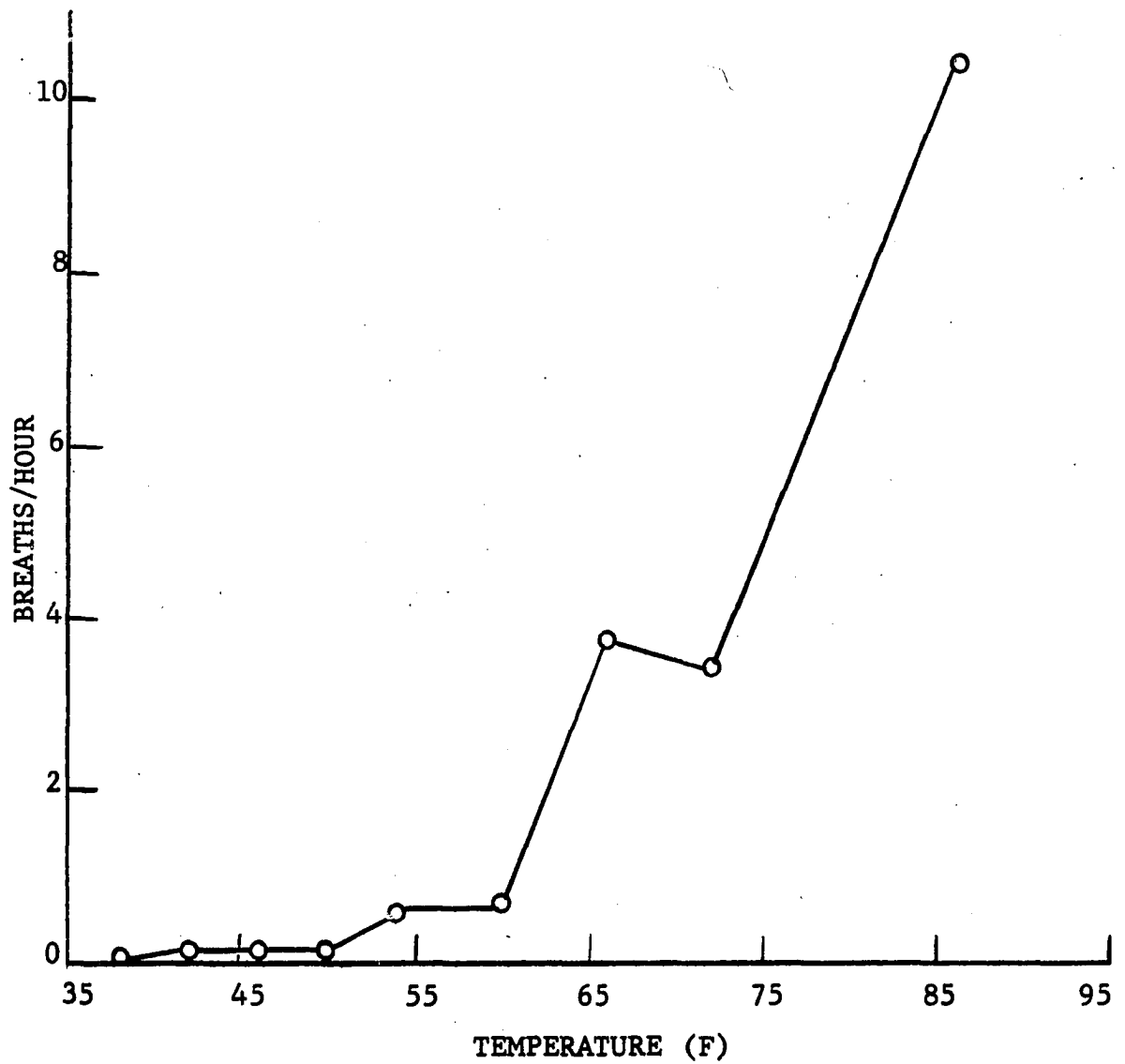


Figure 7. Rate of aerial breathing of longnose gar C at different temperatures.

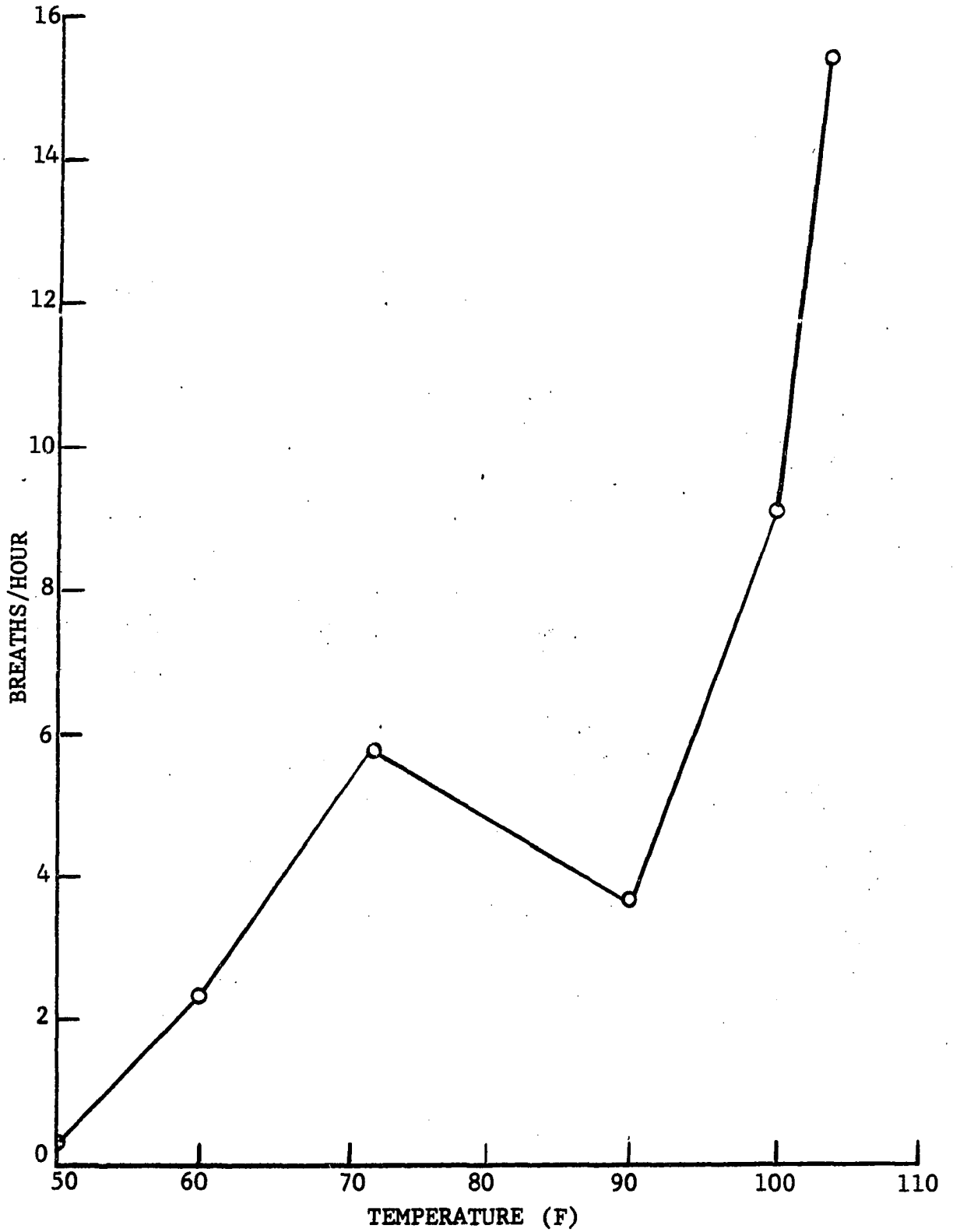


Figure 8. Rate of aerial breathing of spotted gar at different temperatures.

external work rather than to the thermal destruction of enzymes. Clausen (1933) measured the oxygen consumption of the black bullhead, Ameiurus melas, and the large-mouth bass, Micropterus salmoides, while increasing water temperature. He found that from 8 to 12 C oxygen consumption of the black bullhead increased rapidly; from 12 to 15 C it decreased markedly; from 15 to 20 C it rose again and declined above 20 C. From 9 to 10 C the oxygen consumption of the large-mouth bass was low; from 10 to 13 C and from 15 to 19 C it increased. He reported that these fluctuations in the oxygen consumption are supposed to be caused by some physiological regulator located probably in the central nervous system, because animals whose central nervous systems were made ineffective did not show this characteristic pattern. Wells (1914), working with Ameiurus melas, Notropis atherinoides, and N. cornutus, reported that in general the increase or decrease of activity proceeds regularly and proportionately with the change in temperature but at the higher and lower limits of temperature there is a breaking over of the bounds of normal activity and a period of abnormal activity ensues. He concluded that these fishes possess a temperature regulating mechanism which is much more delicate than that of warmer animals, though not as efficient in maintaining a

constant body temperature. Sullivan and Fisher (1947), working with brook trout, Salvelinus fontinalis, found that the number of movements increased with the increase in temperature until a maximum was reached at a temperature considerably below the lethal one. With further increase in temperature the number of movements decreased. Graham (1949) also measured the effects of temperature on the metabolism of brook trout, Salvelinus fontinalis, and found that the active rate of the oxygen uptake increased with increasing temperatures up to approximately 19 C above which it declined. Sullivan and Fisher (in Sullivan, 1954) reported that in experiments where fish (species unnamed) were allowed to equilibrate for a period of twenty minutes to a changed temperature the frequency of their movements was least at low temperatures and increased with the increase in temperature to a peak which occurred at the temperature normally selected by the same animal in a gradient; it then decreased as temperature was further raised, reaching a second low at a temperature several degrees below the lethal level. With further increase in the temperature, frequency was increased again rapidly as the fish died. This relation was found to be the same whether temperatures were changed in regular sequence or at random.

One environmental factor, the dissolved oxygen, was not measured and probably supplemented the effect of lowered metabolism at lower temperatures. Riggs and Moore (unpublished) showed that gars permitted to breathe air could live at least 80 hours in water in which there was no measurable dissolved oxygen, but died quickly in oxygen saturated water if prevented from breathing air.

CHAPTER IV

EFFECT OF LIGHT

Procedure

Both longnose and spotted gars were used in the experiments designed to determine whether the rate of aerial breathing is affected by the presence or absence of light when other factors are kept constant. These experiments were conducted in the same room in which temperature was tested, and since the experiments to determine the effects of temperature, described in the previous chapter, were performed in both light and darkness, the data simultaneously provided information on the effects of light.

Results

Data for the longnose gar (Table 2), on inspection alone, clearly show that at lower temperatures (38 to 54 F) the rate of aerial breathing was slightly higher during the day than at night but at higher temperatures (above 54 F)

they breathed more during the night than the day. There are many methods by which the data for the light experiments of the longnose gar could have been analyzed. A 2 x 2 contingency test (Table 3) was simple and satisfactory and was employed to show that the difference in the aerial breathing rates during day vs. night was statistically significant. Although the spotted gar was not tested below 50 F, the data (Table 2) for higher temperatures (above 50 F) show that, in general, they too had the tendency of higher breathing rate during the night than in the day.

In an attempt to determine whether the breathing rate of gars is affected strictly by presence or absence of light rather than natural day and night regardless of the amount of light, another experiment was set up in which the light was kept off during day (6:30 A.M. to 6:30 P.M.) and on during night (6:30 P.M. to 6:30 A.M.). The temperature was kept constant at 72 F and a continuous recording of breathing was made. The results of this experiment with both longnose and spotted gars (Table 4) showed a tendency for the breathing rate to be slightly greater in the dark than in light although this difference was statistically significant for only one fish (longnose gar C).

In one additional experiment, conducted at 72 F, the

TABLE 2

Number of aerial breaths during successive periods of day
(6:30 A.M. to 6:30 P.M.) and night (6:30 P.M. to 6:30 A.M.)

Temp.	Trial		Trial		Trial		Trial		Trial		Trial		Trial	
F	1	2	3	4	5	6	7							
Longnose Gar A														
	D*	N**	D	N	D	N	D	N	D	N	D	N	D	N
38	1=	1	0=	0	1>	0	0=	0	0=	0	1=	1	0=	0
42	1>	0	1>	0	0=	0	1=	1	1=	1	1>	0		
46	3>	2	1>	0	2=	2	3>	2	2>	0	0=	0		
54	11<	30	10<	23	4<	9	2<	5						
66	21<	46	57<	61	10<	53								
Longnose Gar B														
38	0=	0	0<	1	0=	0	0=	0	0=	0	0=	0	0=	0
42	1>	0	0=	0	0<	2	1=	1	1>	0	1>	0		
46	0=	0	1>	0	2<	3	2>	0	4>	0				
50	1>	0	0=	0	3>	2	0<	1	1>	0	1>	0		
54	12>	8	5>	3	4>	0	7>	2						
60	28>	15	36>	23	24<	27	28>	23	31>	28	32<	34	29<	33
66	76<	114	77>	70										
72	16<	72	25<	95	25<	86	83>	81	65<	66	61<	63		
86	153>	148	116<	163	132>	128	103<	117						
96	91<	93	90<	109	83<	84	77<	85						
Longnose Gar C														
38	2>	0	1>	0	0=	0	1=	1	0=	0	2>	1	0=	0
42	1<	2	1<	2	1=	1	5>	0	4>	2	4>	1		
46	1=	1	2>	1	3>	1	3>	0	3>	1				
50	3>	0	1<	3	2=	2	2>	1	2>	1	2=	2		
54	12>	8	11>	3	13>	4	5>	3						
60	8<	11	12>	9	6<	8	7>	5	8=	8	9>	6		
66	45<	66	29<	49	30<	48								
72	24<	114	10<	80	11<	36	46<	59	45<	51	11>	7		
86	148<	158	126<	142	107<	126	92<	103						
Spotted Gar														
50	4<	6	3=	3	3>	0	2<	4	3<	4	1<	3		
60	42<	49	43>	36	42<	51	14<	18	21>	12	14=	14	11<	29
72	102<	109	71<	85	64<	83	47<	60	36<	65	49<	64		
90	45<	53	42<	47	45>	42								
100	116<	137	100<	115	120>	103	89<	97						
104	195>	175												

*D = Day; **N = Night.

TABLE 3

2 x 2 Contingency-test comparison of data from Table 2

Longnose Gar A				
Temp. Range F	D > N*	D < N	Total	
38 - 46	8.0 (4.26)	0.0 (3.74)**	8.0	Chi-square (corrected for continuity) with 1 degree of freedom = 11.30 .001 > P > .0005
54 - 66	0.0 (3.74)	7.0 (3.26)	7.0	
Total	8.0	7.0	15.0	
Longnose Gar B				
Temp. Range F	D > N	D < N	Total	
38 - 54	14.0 (9.66)	4.0 (8.34)	18.0	Chi-square (corrected for continuity) with 1 degree of freedom = 5.87 .025 > P > .01
60 - 96	8.0 (12.34)	15.0 (10.66)	23.0	
Total	22.0	19.0	41.0	
Longnose Gar C				
Temp. Range F	D > N	D < N	Total	
38 - 54	18.0 (11.58)	2.0 (8.42)	20.0	Chi-square (corrected for continuity) with 1 degree of freedom = 15.17 P = >.0005
60 - 86	4.0 (10.42)	14.0 (7.58)	18.0	
Total	22.0	16.0	38.0	

*D = Day; N = Night.

**Calculated expected values for the 2 x 2 contingency analysis are shown in parenthesis below the observed values.

light was turned on and off continually at 2-hour intervals for six days and the breathing rate of both longnose and spotted gars was continuously recorded. These data were analyzed separately for the light and dark periods (Table 5). Once again both species showed a tendency for breathing more in darkness than in light, although this difference, when analyzed by both parametric and non-parametric tests, came out to be significant only in the single spotted gar and in one of the longnose gar (fish C).

The results of these experiments indicate that at 72 F both longnose and spotted gars have a higher rate of aerial breathing during darkness than in light irrespective of the fact that darkness occurs in natural day or night time. It can also be generalized from the data that both species of gars have higher breathing rates during day at lower temperatures (38 to 54 F) and during darkness at higher temperatures (above 54 F).

Discussion

Light is also known to be among the several modifying factors which have been shown to affect metabolism in animals (Prosser and Brown, 1961). Numerous studies, on the diurnal activity rhythms in fishes, have been published.

TABLE 4

A comparison of rates of aerial breathing in light vs. darkness when the light was off during the day (6:30 A.M. to 6:30 P.M.) and on during the night (6:30 P.M. to 6:30 A.M.)

Species and Specimen	Breaths/Hour		Number of Trials*	t	Degrees of Freedom	Paired-t (2-tailed P)	Non-Parametric Sign Test (2-tailed P)
	Light	Dark					
Longnose Gar B	5.471	7.096	6	1.787	5	.2>P>.1	0.469
Longnose Gar C	0.221	3.624	"	-8.003	"	P=<.01**	0.031**
Spotted Gar	3.333	4.416	"	-1.565	"	.2>P>.1	0.187

*Trial = 24 hours.

**Significant at 5% level.

TABLE 5

A comparison of rates of aerial breathing in light vs. darkness when the light was alternately on and off at 2-hour intervals

Species and Specimen	Breaths/Hour		Number of Trials*	t	Degrees of Freedom	Paired-t (2-tailed P)	Non-Parametric Sign Test (2-tailed P)
	Light	Dark					
Longnose Gar B	5.249	6.777	6	2.667	5	.05>P>.02	0.469
Longnose Gar C	0.458	2.527	"	-43.845	"	P=<.01**	0.031**
Spotted Gar	2.527	5.472	"	- 6.872	"	P=<.01**	0.031**

*Trial = 24 hours.

**Significant at 5% level.

Sullivan and Fisher (1947) found that the brook trout, Salvelinus fontinalis, avoids light in preference to darkness and selects temperature more sharply in dim light than in bright light. Spencer (1939) reported that the pumpkinseed sunfish, Lepomis gibbosus, is more active in day, whereas carp, Cyprinus carpio, and minnow, Umbra limi, and rock bass, Ambloplites rupestris, are more active at night.

CHAPTER V

EFFECT OF FEEDING

Procedure

The experiments designed to test the effect of varying amounts of food on the rate of aerial breathing of the longnose gar were conducted at 72 F. The only two specimens (fish B and C) available at the time were used in the feeding experiments. Since it was necessary to know the exact time that the fish ate, a method was designed whereby the experimental fish could be fed the desired amount of food at any designated time. A narrow, hollow, glass rod about two feet long was threaded with a flexible steel wire. One end of this wire was bent into a hook on which a freshly killed mosquitofish, Gambusia affinis, was impaled. This impaled fish was suspended beside the snout of the gar and with a little jiggling of the rod it was quickly seized. After the mosquitofish was thus seized by the gar, the rod was withdrawn for feeding additional fish if so desired. They

normally would not have eaten these dead fish unless some movement was effected. This method of feeding the gar proved to be very successful. Only the mosquitofish, Gambusia affinis, of 4.0 cm average total length, were used as food and throughout these experiments the lights were on from 6:30 A.M. to 6:30 P.M. and off from 6:30 P.M. to 6:30 A.M.

Before the start of the feeding experiments, long-nose gars were starved for one week and their breathing rate was recorded with the Physiograph for two days prior to feeding. Successive recordings of their breathing were made when they were fed every 24 hours, one, two, and four mosquitofish in three separate experiments respectively. Finally after-feeding recordings of the breathing rates were made when the gar were fed as many fish as they could eat beginning at 10:00 A.M. every day. In this experiment each gar ate an average of 14 mosquitofish per day.

Result

The data show (Table 6 and Figure 9) that at 72 F the breathing rate decreased during starvation below that when they are fully fed. After starvation the breathing rate of the longnose gar B and C was 3.062 and 2.291 breaths/hour respectively (Table 8) compared with 5.124 and 3.430 breaths/

hour (Table 1) for the same fish when they were regularly fed. Also, the aerial breathing rate increased with the increase in the intake of food and reached a maximum when the fish were fed to satiation.

Discussion

Respiration in animals is affected by both the amount of food eaten and starvation. Smith (1935a, 1935b) measured the rate of metabolism of starved African lungfish, Protopterus aethiopicus, and found that beginning with the first day of fasting, the oxygen consumption fell in a regular manner for at least the first 300 days, at the end of this time reaching levels of 10 to 20 per cent of oxygen consumption of the fully-fed fish. In another experiment he measured the metabolic rate of well-nourished lungfish fed meat at decreasing intervals, or daily in increasing quantities, and found that the oxygen consumption reached a constant maximum level when the caloric intake was slightly in excess of that required to produce caloric equilibrium.

The feeding behavior of gar has been known for a long time (Agassiz, 1859). It is fascinating to watch young gars feeding; their behavior is quite different from that of the majority of the other fishes. Most piscivorous fishes take

and immediately swallow their food. Young gars are piscivorous and apparently feed only on small live fishes and other live aquatic organisms. In the laboratories at the University of Oklahoma Biological Station and the Norman campus they have never been observed to eat dead fish except the freshly killed ones offered to them in the manner already described. Older gars readily eat both live as well as dead fish. Young gars approach their prey slowly from the side and catch it with a quick sideways motion of the elongated jaws. They hold their prey crosswise between the jaws and by a series of biting movements often shift it back and forth, finally bringing it into the proper position for swallowing head first. Once the prey is turned in swallowing position the actual act of swallowing takes only a fraction of a second. Sometimes the prey is carried about for several minutes, however, before it is turned to swallowing position. Active pursuit of prey that involves strenuous activity or that should create sudden stringent oxygen demands has not been observed.

Not much physical activity of gars was involved in the feeding experiments as they were artificially fed. The increased rate of aerial breathing after feeding was probably caused by the increased physiological demand produced by digestion. It was obvious, however, that during the feeding

experiments these fish learned quickly that they were to be fed. As soon as I entered the room they would swim to the end of the aquarium where they were fed and wait until the food was presented.

TABLE 6

Effect of varying amounts of food on the rate of aerial breathing of
longnose gar at 72 F

	Starvation	1 mosquitofish	2 mosquitofish	4 mosquitofish	Feeding to Satiation
Fish B (Average) (Breaths/Hour)	3.062	3.676	4.502	8.271	15.995
Fish C (Average) (Breaths/Hour)	2.291	4.228	4.708	5.941	10.906
Number of Trials*	2	4	4	4	4

*Each trial consisted of 24 hours of recording.

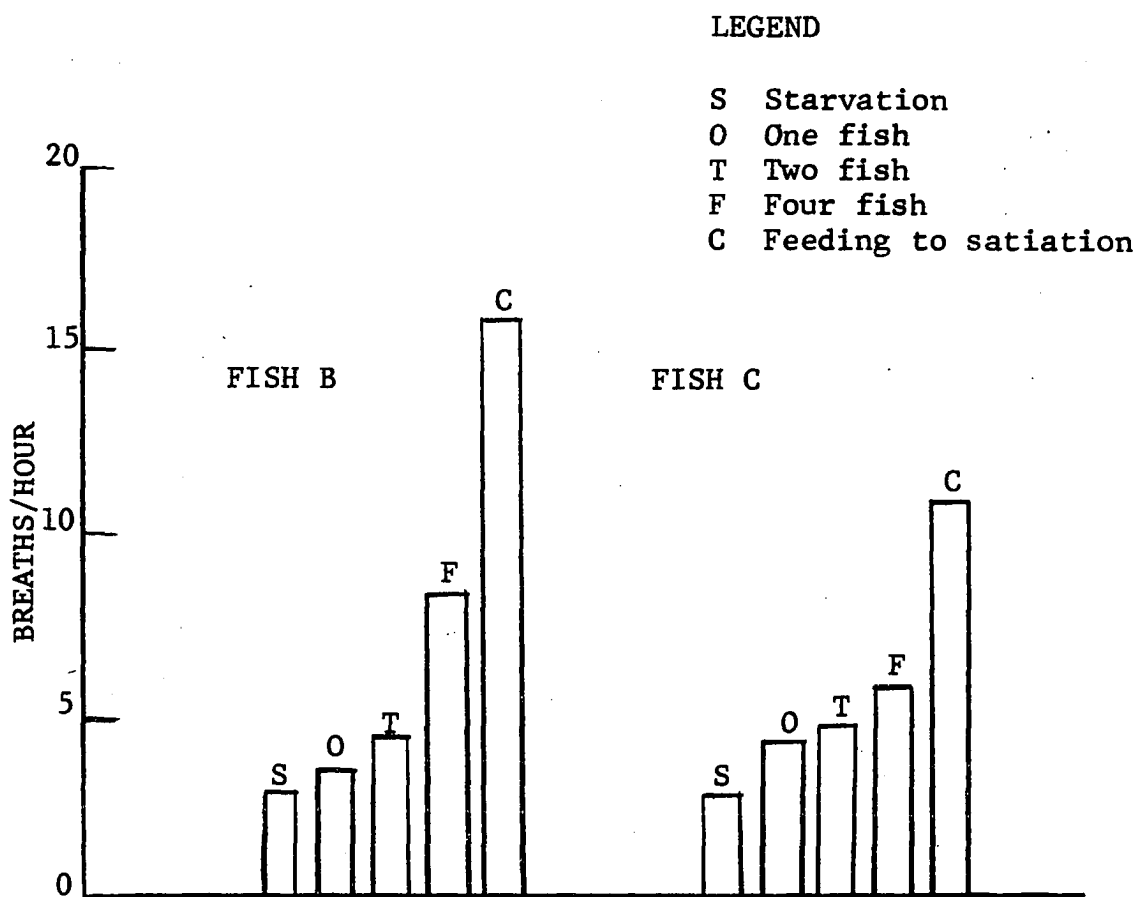


Figure 9. Comparison of breathing rates of the longnose gars B and C at 72 F after eating different quantities of food.

CHAPTER VI

EFFECT OF ACTIVITY

Procedure

Two specimens of the longnose gar (fish B and C), were used to determine the effect of activity on their rate of aerial breathing. All experiments were conducted at 72 F during the day with the lights on. Simultaneously two fish were made to swim continuously for intervals of one-half hour by touching them with a meter stick when they attempted to stop. Immediately after the termination of this induced period of activity the breathing rate was recorded with the Physiograph and the first interval between breaths measured was the one immediately following the first recorded breath.

Results

The rate of aerial breathing for both fish was very rapid immediately after activity, and later decreased to the normal breathing rate. The intervals between successive

breaths immediately after activity were obviously much shorter than "normal" and lengthened as time passed (Figures 10 and 11; Tables 7 and 8). The data show that fish B took about 45 minutes and fish C about 56 minutes before the breathing rate returned to normal. and that during this recovery period they took an average of 8 and 10 breaths respectively. Based on the data taken during the day at 72 F in the temperature experiment (Table 1), the average time between successive breaths for longnose gars B and C was 15 and 30 minutes respectively. From the data obtained in the activity experiment (Tables 7 and 8) it is obvious that at 72 F increased activity increases the rate of aerial breathing of the longnose gar above that carried on during normal activity.

Discussion

It is common knowledge that increased activity results in increased oxygen demand by the organism involved. There are several reports in the literature that fishes consume relatively more oxygen during periods of activity than during quiescence. Higginbotham (1947), working with Schilbeodes nocturnus, and Ictalurus lacustris punctatus (sic), found that these fishes used oxygen at consistently

TABLE 7

Time between successive breaths of longnose gar B immediately after activation at 72 F

Sequence of Breaths	Trials							Average (Min.)
	1 (Min.)	2 (Min.)	3 (Min.)	4 (Min.)	5 (Min.)	6 (Min.)	7 (Min.)	
2	6	1	1	2	3	9	8	4.285
3	7	2	1	6	7	5	7	5.000
4	6	10	1	3	4	3	4	4.428
5	3	3	2	3	2	9	1	3.285
6	16	4	9	1	3	4	4	5.857
7	12	9	18	1	1	32	6	11.285
8	17	2	30	7	2	15	6	11.285
9	24	7	29	3	11	24	1	14.142
10	20	6	22	7	3	28	47	19.000
11	20	4	24	7	4	24	36	17.000
12	23	3	29	1	1	28	32	16.713
13		24	25	25	29	25	25	25.500
14		8	20	13	14	26	51	22.000
15		10	28	24	22			21.000
16		12	33	19	13			19.250
17		14	24	13	7			14.500
18		24	29	27	5			21.250
19		12	22	21	34			22.250

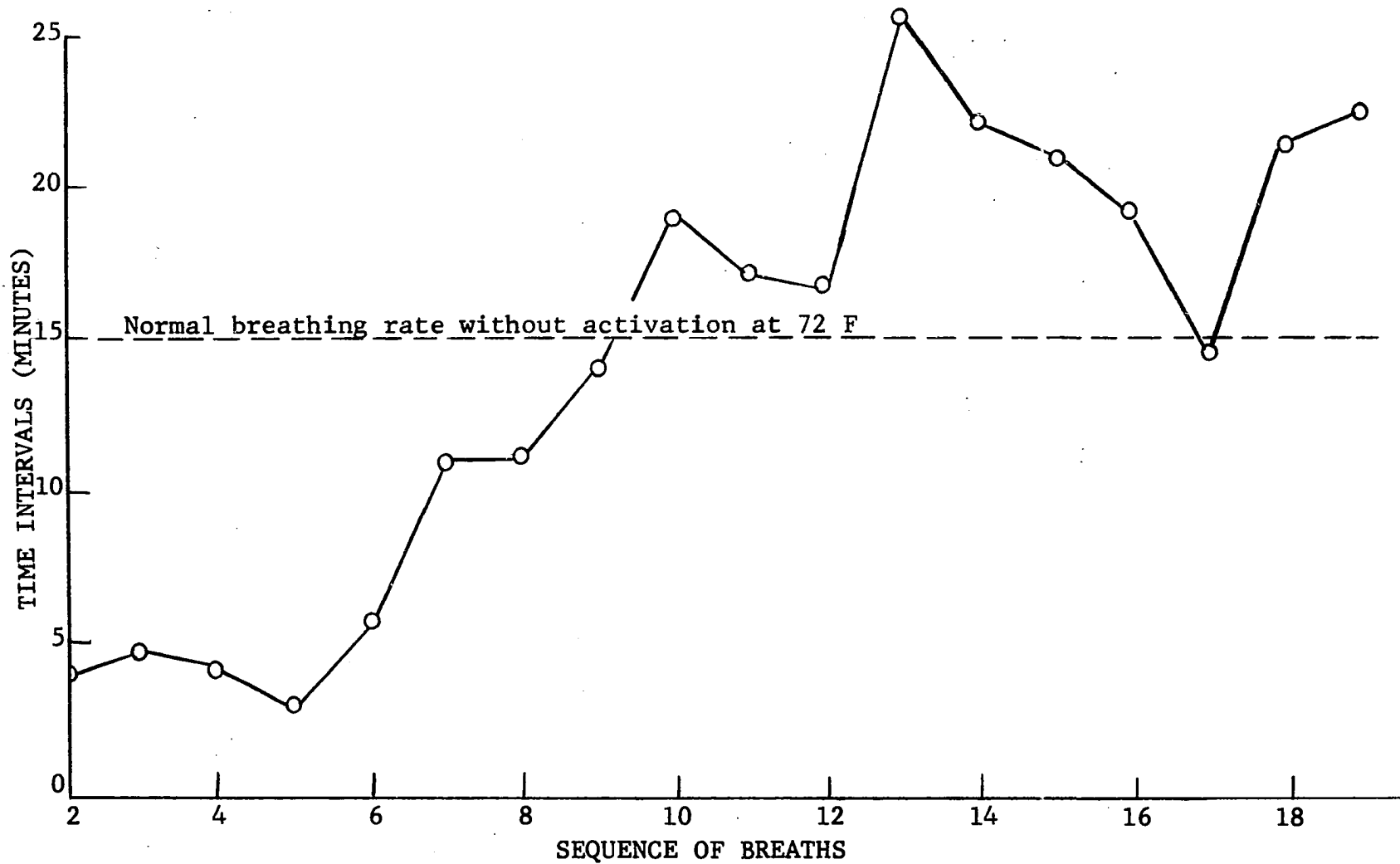


Figure 10. Average intervals between successive breaths of longnose gar B at 72 F, immediately following a half-hour period of activation.

TABLE 8

Time between successive breaths of longnose gar C immediately after
activation at 72 F

Sequence of Breaths	<u>Trials</u>					Average (Min.)
	1 (Min.)	2 (Min.)	3 (Min.)	4 (Min.)	5 (Min.)	
2	6	1	2	2	2	2.600
3	1	1	2	3	2	1.800
4	8	1	1	6	1	3.400
5	15	1	5	1	3	5.000
6	12	1	8	6	11	7.600
7	20	3	8	1	8	8.000
8	20	1	11	2	5	7.800
9	12	2	6	3	23	9.200
10	14	1	1	6	30	10.400
11	36	6	34	19	22	23.400
12	36	2	21	19	30	21.600
13	20	11	28	39	43	28.200
14	22	7	27	34	30	24.000
15	37	17	24	17	28	24.600
16	24		27	34		28.333
17	38		25			31.500

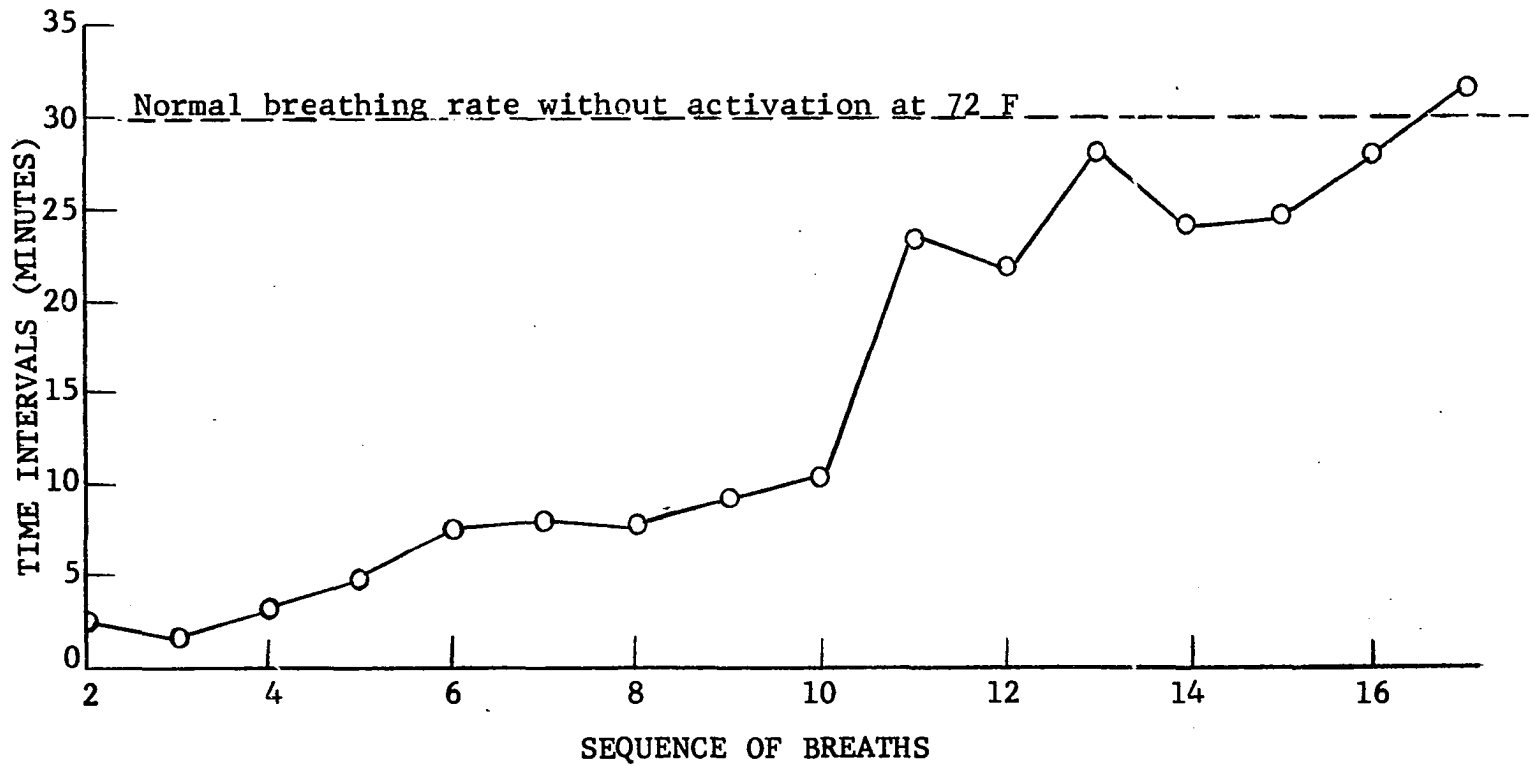


Figure 11. Average intervals between successive breaths of longnose gar C at 72 F, immediately following a half-hour period of activation.

higher rates during the late afternoon when they were more active. Spoor (1946) showed a close relationship between activity and oxygen consumption in the goldfish.

From my observations and those made at the University of Oklahoma Biological Station, gars are lethargic fishes in the laboratory and in the lacustrine habitat. They spend much time either resting on the bottom or suspended, practically motionless, just beneath the surface of the water. Even when hunting or pursuing food their activity seems to be limited to the least amount of effort necessary. Therefore, it seems probable that activity would normally play a lesser role than temperature and feeding in affecting aerial breathing. In the lotic environment, however, activity has been observed to be much greater and probably has a greater effect on aerial breathing.

Based on these activity experiments and the general observations which I made throughout the study it seems obvious that gars quickly acclimate to laboratory conditions. Entrance to the lighted room or my presence in the room, close to or away from the aquaria, had no apparent effect on their activity. Only when I entered the dark room and turned on the light, and during the course of the feeding experiments, did my presence arouse any change in activity which could be determined by observations or the recordings.

CHAPTER VII

GENERAL DISCUSSION

Although there are several extrinsic and intrinsic factors which influence respiration in animals, I tested only four (temperature, light, feeding, and activity) for their possible influence on the rate of aerial breathing in Lepisosteus. Temperature not only affected the rate of aerial breathing, but also activity (swimming) and appetite. In general the rate of aerial breathing increased with the increase in temperature, although the increase was neither uniform nor consistent. Although my data do establish that the rate of aerial respiration for both the longnose and spotted gars, like that of the aquatic respiration for several other fishes (e.g., goldfish, black bullhead, and largemouth bass), temporarily decreases at one or two points as the temperature increases, they do not establish a reason for these decreases. Knowledge of the causes of these periods of rate decrease might well contribute to the explanation

of the irregularities in rate of air breathing mentioned earlier.

Many studies have proved that the metabolic rate of poikilothermic animals is directly and positively affected by temperature, and that an increased metabolic rate generally results in an increase in the oxygen demand of the animal. The occurrence of air-breathing organs is accompanied by reduction of the gills (Carter, 1957). If this is true for gars, it is probable that the oxygen demand at temperatures below approximately 50 F is satisfied by gill respiration, and above this temperature the fish is increasingly more dependent upon aerial respiration. The fact that more oxygen can be dissolved in colder water must also affect this situation.

The literature includes a number of reports that photoperiodism in many fishes affects their pattern of activity. At least in the laboratory, gars appear more active at night than during the day, and may well be so adapted. Thus my data which indicate that at higher temperatures (above 60 F for spotted gar and 54 F for longnose gar) gars have a greater rate of aerial breathing during night than day might be explained by the fact that gars are more active at night than in the day, and thus the effect of light is an indirect

one rather than a direct one.

When the other environmental factors (temperature, light, and feeding) remained constant, induced activity resulted in a marked increase in rate of aerial breathing. This is apparently a direct effect brought on by immediate increased oxygen demand. The period of recovery to the normal rate of air breathing was short.

The ingestion of food was immediately followed by an increase in rate of aerial breathing. Furthermore, the increase in breathing rate was directly proportional to the amount of food that was eaten. In the natural environment feeding should produce an even more marked increase in aerial breathing rate since the physical activity involved in hunting and capturing the prey and the nervous activity which results from the excitement brought on by the awareness of the presence of food should supplement the increased oxygen demand resulting from the physiological activity of digestion.

The data of this study clearly indicate that all of the four factors tested affect the rate of aerial breathing of gar in the same general way that air breathing of tetrapods is affected. These data are important for this reason if for no other and also that they substantiate the existing evidence that gars are air breathers, that their lung or

swim bladder is a respiratory organ, and that aerial breathing is of vital physiological importance to them. It is also possible that the data provide evidence, though inconclusive, which might explain to some extent the irregular rate of aerial breathing exhibited by these fishes.

CHAPTER VIII

SUMMARY

1. Studies were made of the effects of temperature, light, feeding, and activity on the rate of aerial breathing of both the longnose gar, Lepisosteus osseus, and the spotted gar, L. oculatus.

2. Continuous written records of aerial breathing under different experimental conditions were made with a Physiograph. A method employing strain-gauges was satisfactory for recording breathing but was not used in this study.

3. Temperature affected aerial breathing rate of gars as well as activity (swimming), and appetite.

4. At the minimum temperature tested (38 F), the aerial breathing rate for the longnose gar was practically zero.

5. Aerial breathing began when the temperature rose above 50 ± 4 F.

6. At the lower (38 to 50 F) and higher (above 90 F)

temperatures the appetite and the swimming activity of the gars declined considerably. The maximum activity and feeding occurred in the range of 66 to 75 F.

7. In general, the rate of aerial breathing increased with the increase in temperature but this increase was neither uniform nor consistent.

8. Both the longnose and the spotted gars died at 104 F; apparently the highest temperature tolerable for these species.

9. Up to about 54 F, the longnose gars had a slightly higher aerial breathing rate during the day than at night, whereas above 54 F they breathed more during night than day. The spotted gar was not tested below 50 F, but above 60 F it too showed a higher breathing rate at night than during day.

10. At 72 F gars had a higher rate of aerial breathing during darkness than in light irrespective of whether darkness occurred in the natural day or night.

11. Feeding increased the rate of aerial breathing of the longnose gars; the greater the amount of food eaten the greater the increase in the aerial breathing rate.

12. The aerial breathing rate of longnose gars was higher when they were regularly fed than during periods of starvation.

13. Induced swimming activity increased the rate of aerial breathing of the longnose gar. After the cessation of the activity the period of recovery back to the normal breathing rate was short.

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APPENDIX TABLE 1

Test of significance between 35- and 50-gallon aquaria

Species	Specimen	Temperature Range F	Condition	t	Degrees of Freedom	Paired-t 2-tailed P	Non-Parametric Sign Test (2-tailed P)	Remark
<u>L. oculatus</u>	A	50-72	Dark	2.069	2	.20>P>.10	0.250	Accept H ₀
"	"	"	Light	1.841	"	.40>P>.20	0.750	"
<u>L. osseus</u>	A	38-66	Dark	-0.446	4	.80>P>.60	0.625	"
"	"	"	Light	-0.535	"	"	"	"
"	B	38-72	Dark	-0.081	7	.60>P>.40	0.437	"
"	"	"	Light	0.993	"	.40>P>.20	0.382	"