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OF THE MILK FISH**

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### INTRODUCTION

SEASONAL variations in temperature, dissolved oxygen and salinity have great influence upon marine life. However, these factors incidental to the tropical environment have not been fully investigated. In this paper some effects of a sudden change in temperature as well as the effects of various partial pressures of oxygen upon its uptake are presented with reference to *Chanos chanos*—the milk fish, so widely used in fish culture. An attempt has also been made to relate this study to a limited range of size of the fish.

A measure of the metabolism or energy turnover of the fish is necessary in order to estimate the requirements for promoting and sustaining growth. Since any change or regulation of metabolism is directly reflected in respiration, the uptake of oxygen by the fish is taken as a measure of its metabolism. Two levels of metabolism are usually recognised: *Active* metabolism which is the total or maximum that the animal is capable of, and *Standard* which is the measurable minimum required for sustaining all ancillary activities of life, below which level the animal is bound to die sooner or later. As established by Fry (1947), the 'scope for activity' of the animal is conditioned by the relation the two metabolic levels bear to each other. This indeed has been demonstrated in various ways by Spoor (1946), Fry and Hart (1948), Graham (1949), and Job (1955); and the importance of relating oxygen consumption precisely to the activity of the fish in similar studies needs no further emphasis. In this investigation, because of limitations in technique the fish could be kept in a routine state of activity only and were obviously not executing any consistently active swimming movements, hence the term 'routine-active' is employed here.

Duodoroff (1942, 1945), Brett (1944, 1952) and Fry (1947) have shown that thermal adaptation both in nature and in experimental conditions is related to the previous temperature history; and that, from this experience the fish acquires various degrees of ability to cope with the temperature conditions of the environment. Brett and Fry have described this effect as

'acclimation' and the term has been used in this paper also, to convey the same idea.

Viswanathan and Tampi (1952), working on the same species measured the oxygen consumption of various sizes and at different temperatures. But the measurements were not related to any consistent size-range at the different temperatures. It was mainly to probe into some unsuspected and interesting aspects of this complex problem and to supplement their data, that this investigation was undertaken.

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#### MATERIAL AND METHODS

Fry of *Chanos chanos* were obtained from tidal creeks in Pamban and were reared in large glass tanks with an adequate supply of tap water of low salinity (0.3‰). They were fed with unlimited quantities of standard food consisting of sheep's liver, clams and seaweeds mixed in definite proportions and ground into a paste. Even though the temperature of water fluctuated over the year, the experiments were so timed that the daily temperature for over a month remained around  $29 \pm 1^\circ \text{C}$ ., before and during the experimental period. Fishes weighing from 0.7 gm. to 11.0 gm. were used in the experiments.

The respiration chambers, Fig. 1, consisted of culture bottles and Ehrlenmeyer flasks, 50 ml. to 5,000 ml. in capacity. The size of the vessel used was such that the enclosed fish was quite free to swim round. The chamber was closed with a rubber stopper carrying both the inlet (1) and the outlet (2). The former was a burette, which by opening the stopcock, would deliver through a long capillary (3) a measured volume of water to the bottom of the chamber. At the time of sampling, the outlet (2) which was normally kept plugged was opened so that an equal volume was simultaneously expelled. Samples were taken at 10, 20, 30 and 60 minute intervals. The chambers were held in water-baths at  $25^\circ \text{C}$ . and  $29^\circ \text{C}$ . and maintained constant to  $\pm 0.2^\circ \text{C}$ ., throughout the duration of the experiment. Unmodified Winkler technique was employed for measuring dissolved oxygen and 5 ml. or 10 ml. samples were usually analysed. The respiration chambers were

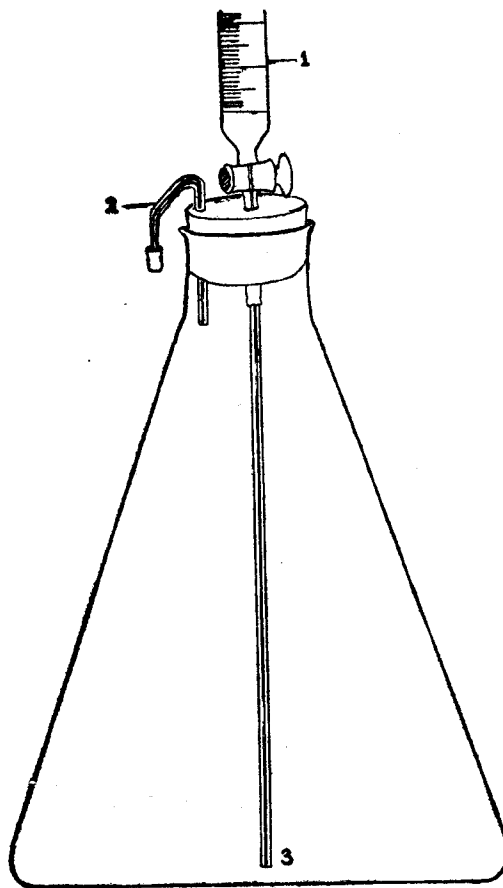


FIG. 1. A simple apparatus for measuring the routine-active oxygen consumption of a fish. 1 and 3. Water inlet. 2. Water outlet.

agitated by the running motor of an air-compressor unit and this kept the fish reasonably active. Some minutes before sampling the motor was turned off so that the fish became more quiescent and did not disturb the water in the chamber while the sample for analysis was actually being drawn. By using coloured water under identical conditions and by repeated tests, the above procedure was adopted so that the water drawn off for sampling did not become contaminated with the water that was introduced, and it represented only the true sample from the chamber.

The pH was measured both at the beginning and at the end of the experiment and the change was always less than 1.0 in the range 8.5 to 7.2. The total volume of water in the chamber was measured and the fish were weighed. The fish were used individually in each experiment. The rate of

oxygen consumption was calculated from the rate of fall in the oxygen level in the sample, with necessary corrections, and expressed in ml./hr. Owing to an unforeseen accident, the stock of fish was lost and the data obtained at 25° C. suffered in consequence and work at other temperatures had to be abandoned. The oxygen consumption data obtained in preliminary experiments showed that *Chanos* is dependent upon the oxygen tension as it obtains under air saturation (approximately 150 mm. Hg) both at 25° C. and 29° C. In order to extend this study into the region of 'non-dependence' (Job, 1955), the oxygen tension was raised to a maximum of 300 mm. Hg. However, unlike the results of Wiebe (1933) and Leiner (1938) there was no reduction in the oxygen uptake under high tensions of the gas. This was probably the effect of keeping the fish uniformly active as has been noted in the speckled trout by Job (1955).

#### RESULTS

The data were plotted on a double logarithmic grid of oxygen consumption against weight (Fig. 2). In Panel A are the data obtained at 29° C. which was both the acclimation and experimental temperature. In Panel B, are the data from fish at 25° C., which were acclimated to 29° C. The partial pressure of oxygen is given at the upper end of each curve. The regression values for the curves in Panel A, from 300 mm. Hg to 25 mm. Hg are :

$$Y = 0.3675 + 0.7952x; \quad Y = 0.2308 + 0.8236x;$$

$$Y = 0.1970 + 0.8242x;$$

$$Y = 0.1567 + 0.8124x; \quad Y = 0.0995 + 0.7765x;$$

$$Y = 0.0058 + 0.6849x; \text{ and}$$

$Y = -0.6181 + 0.7746x$ , respectively. The corresponding values for Panel B are:

$$Y = -0.0034 + 0.9573x; \quad Y = -0.2521 + 1.0615x;$$

$$Y = -0.3452 + 1.0842x;$$

$$Y = -0.4895 + 1.1310x; \quad Y = -0.7141 + 1.2003x;$$

$$Y = -0.9651 + 1.2358x; \text{ and}$$

$Y = -1.0258 + 0.8460x$ , respectively. These values were derived from the formula  $Y = bx^k$ , where  $Y$  is the log. rate of oxygen consumption,  $x$  the log. of weight,  $b$  the slope of the line and  $k$ , a constant.

## DISCUSSION

The data in Fig. 2 when replotted and illustrated as in Figs. 3 and 4 bring out clearly the relationship of size to two different temperatures and to various partial pressures of oxygen in the medium respectively.

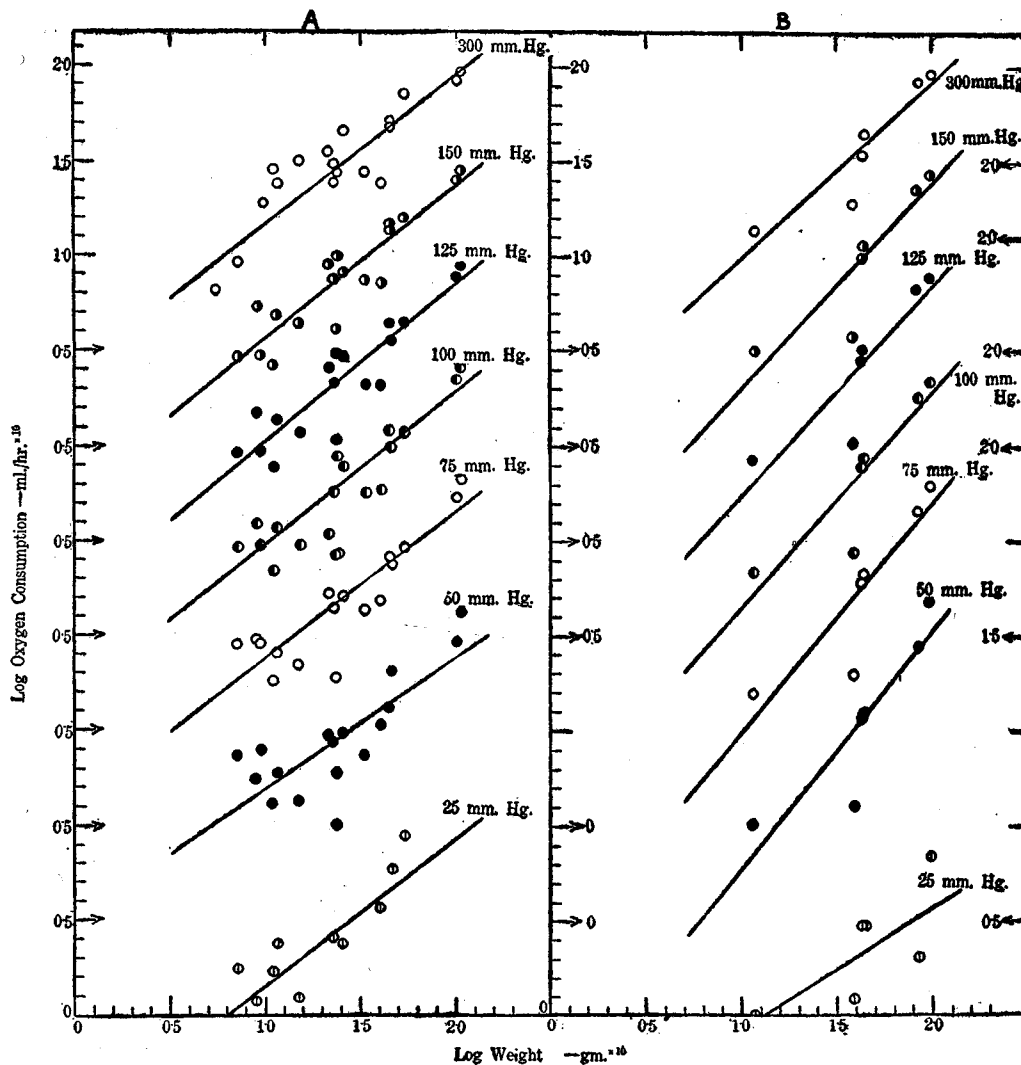


FIG. 2. Data of routine active metabolism at 29° C. (Panel A) and at 25° C. (Panel B). The partial pressures of oxygen are indicated at the upper end of each line. The lines of best fit were drawn according to the principle of least squares. The arrows along the ordinate indicate in this double logarithmic plot, the corresponding cycle of the line immediately opposite.

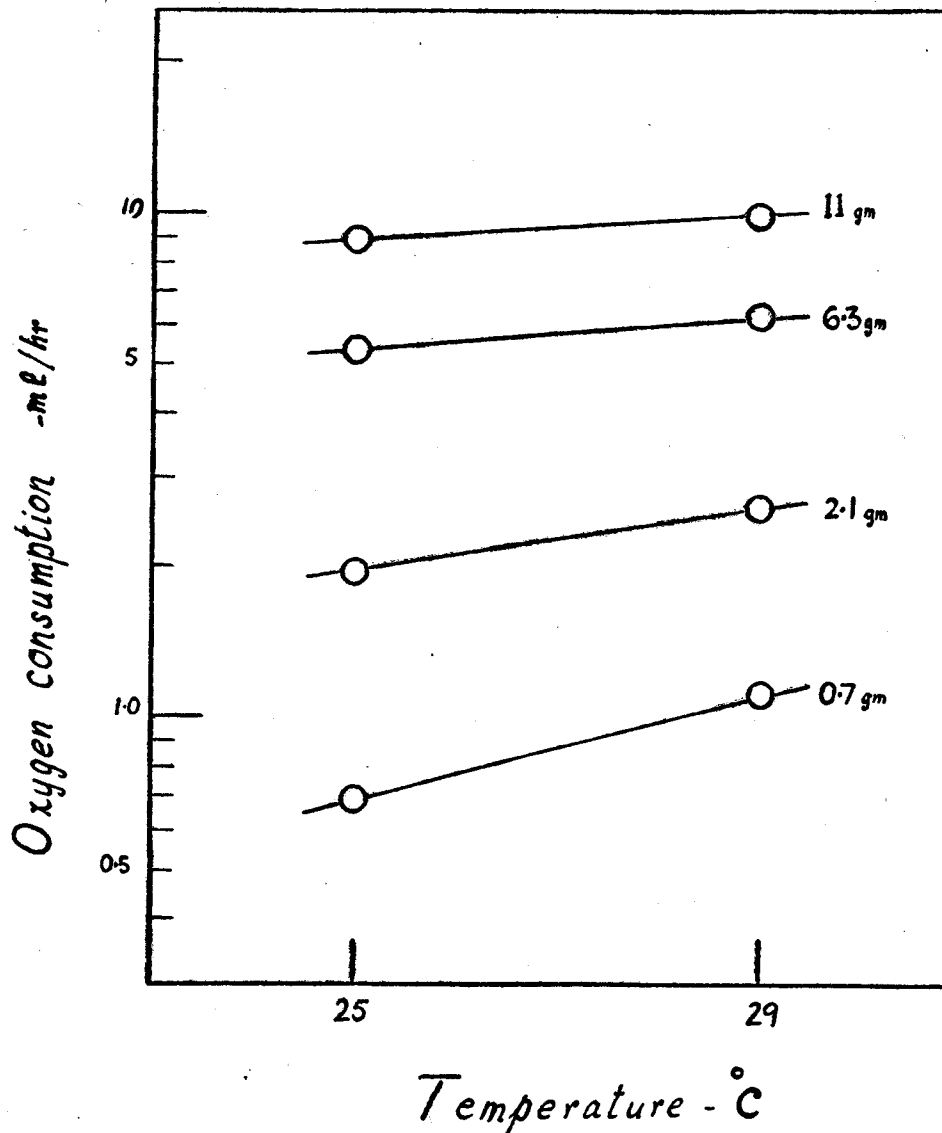


FIG. 3. Illustration of size and temperature relationship in 'routine-active metabolism' at an average  $pO_2$  of 300 mm. Hg. At the upper end of each line is given the respective weights of the fish which were taken in logarithmic intervals except for the last group (11 gm.).

#### *Size and Temperature*

The data obtained for the milk fish bear out in a limited range of size and temperature, the trend obtained in experiments on the speckled trout (Job, 1955), wherein the value of the gradients increased as the temperature

decreased from 20° C. to 5° C. A conspicuous rise in the regression-gradient was obtained at 25° C. compared with those at 29° C. in this work: the former gradients ranging from 0.8460 to 1.2358 and the latter from 0.6849 to 0.8242.

There is, further, a remarkable 'acclimation-effect': the relative response in the metabolic rate at 29° C. (Fig. 2, Panel A) has a regularity which however is not true of the set at the unacclimated temperature of 25° C. (Panel B). Even though the difference is only 4° C., the lower temperature because of the suddenness of impact upon fish acclimated to the higher temperature, acts probably as an accessory factor producing various effects on the different sizes (Table I). In studies of this nature therefore, the previous history of the organism in relation to any particular factor of the environment under investigation cannot be overlooked. Comparison of data obtained at various arbitrary temperatures (Viswanathan and Tampi, 1952) will not therefore give reliable pictures of the metabolic responses of the various sizes.

TABLE I  
*Oxygen Consumption ml/hr.*

Weight gm.	Partial Pressure of Oxygen mm. Hg						
	25	50	75	100	125	150	300
<i>At 29° C.</i>							
0.7	0.11	0.38	0.57	0.71	0.78	0.85	1.11
2.1	0.26	0.82	1.34	1.73	1.93	2.11	2.60
6.3	0.60	1.74	3.16	4.17	4.84	5.13	6.22
11.0	0.91	1.40	4.84	6.50	7.69	8.07	9.84
<i>At 25° C.</i>							
0.7	0.07	0.12	0.20	0.30	0.37	0.44	0.69
2.1	0.14	0.46	0.74	1.00	1.23	1.41	1.95
6.3	0.27	1.82	2.82	3.55	4.07	4.57	5.37
11.0	0.39	3.55	5.37	6.61	7.41	8.13	8.91



At 25° C., while there is a slight fall in the over-all metabolic rates compared with those at 29° C. (Table I), for all sizes, the smallest fish shows the greatest change in the rate of uptake between the two temperatures (Fig. 3). Sumner and Duodoroff (1938) report that reversal of metabolic response takes place when fish acclimatized to high and low temperatures are tested at some intermediate temperature. They state further that heat-resistance did not show any size relationship. This is also largely true in the Pacific salmon as was demonstrated by Brett (1952). But he established that death from low temperature (30% to 50% mortality) was significantly higher in the smaller fish. He suggested that this was due to the greater susceptibility of smaller fish to 'rapid' cold death. The data on the speckled trout (Job, 1955) gave a different view of this temperature-response in fish according to size. Though tentative, it was demonstrated that the 'maintenance costs', that is the % of standard over active metabolism, was relatively higher for the smaller fish than for the bigger fish at the lower temperature. In the milk fish it seems highly probable that the greater dependence of the smallest fish, even at 300 mm. Hg  $pO_2$  (Fig. 4), is due to the higher maintenance costs at the lower temperature and this probably also accounts for the difference in response by size noted by Brett in the Pacific salmon. In the absence of exact measurements of standard metabolic rates for the milk fish this explanation is offered with reservation.

#### *Size and Oxygen Tension*

Another point of interest can also be presented by tracing the relation of size to the ability of the fish to utilise oxygen and the oxygen tension in the medium. As can be seen clearly in Fig. 4, Panel A, when the acclimation temperature and the experimental temperature are identical, the relative ability of the fish to utilise oxygen is the same irrespective of size. The curve for the 11 gm. fish when reduced and superimposed upon that of the 0.7 gm. fish coincides with it along almost the entire range, thereby indicating the similarity in the capacity for oxygen utilisation under all partial pressures. This was also noted in the speckled trout (Job, 1955), and over a wider range in size and temperature. When an accessory factor is introduced, such as in this case a sudden fall in temperature, this relationship does not hold good, as can be seen from Panel B, in the same figure. In the former case (Panel A) the difference in the % of fall in the rate of oxygen uptake at the highest and lowest partial pressures of the gas, between the 11 gm. and 0.7 gm. fish, was 0.66%. In the latter case (Panel B) the corresponding fall was greater, amounting to 5.77%.

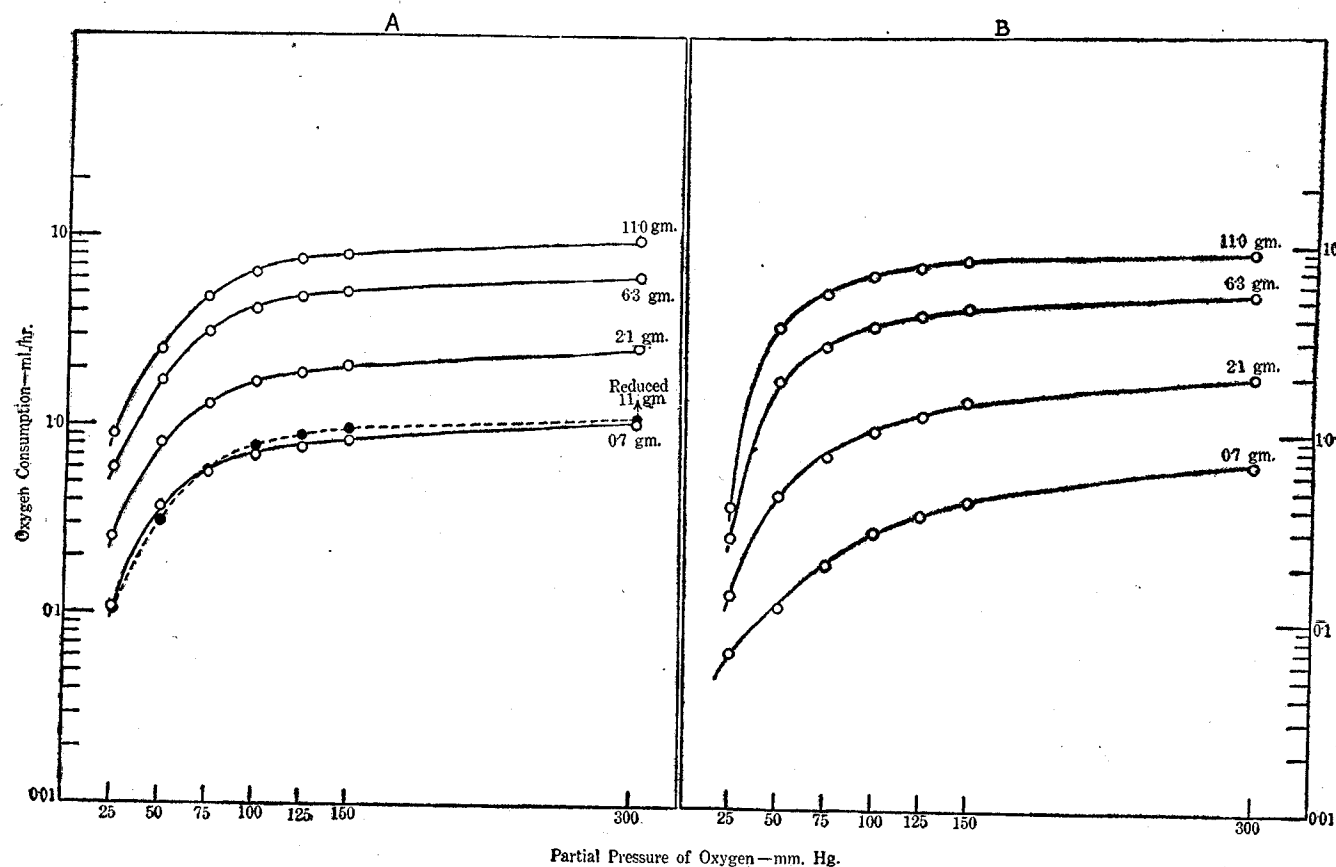


FIG. 4. Illustration of size and dependent respiration at various partial pressures of oxygen. Panel A is the data at 29° C., which was also the acclimation temperature. Panel B is the data at 25° C. The weights of the fish are given at the upper end of each curve. The broken line in Panel A represents the curve of the 11 gm. fish reduced and drawn over that of the 0.7 gm. fish.

### Critical Tension

It will be noted from Fig. 4 that the dependency upon oxygen tension generally becomes very pronounced at nearly 150 mm. Hg  $pO_2$ , and at almost air-saturation levels, as the curve below this point shows a steep fall. In two species of Chironomids inhabiting a pool where oxygen tension was low and ranged from 3 mm. Hg to 92 mm. Hg, Walshe (1948) found that the critical tension was 35 to 65 mm. Hg. Maloeuf (1937), on the other hand, found that a mussel showed dependency even at pressures four times that of air-saturation. Thus there appears to be various kinds of dependency upon the oxygen tension of the medium in different animals. In the milk fish as well as in the speckled trout (Job, 1955), the critical tension was normally at air-saturation pressures. It seems, therefore, that the critical tension itself is an indication of the intensity of normal metabolic activity; and because of its wide variation, a definition of the critical tension with reference to its range and magnitude appears to have its drawbacks. According to Hayes *et al.* (1951), the critical tension is that which is half maximal and which may extend from air saturation to almost zero. From a comparison of curves for the 0.7 gm. fish in Panels A and B in Fig. 4, such a definition seems untenable, even within the same species.

### Oxygen Consumption and Live-fish Transport

In experiments upon milk fish it was found that a 0.7 gm. fish consumed under routine-activity, about 1.1 ml./hr., and a 11.0 gm. fish about 9.84 ml./hr. at 29° C. This was reduced at 25° C. to 0.69 and 8.91 ml./hr., respectively. At the same time, the data obtained at the asphyxial level of oxygen which, however, did not indicate any 'size-effect', either at 25° C. or at 29° C., showed that the average value was 1.05 ml./litre, at 29° C. and 0.78 ml./litre, at 25° C. While it is possible to make use of the former set of data in estimating the maximum oxygen requirements of the particular size of fish during transport, it is not possible to make use of the latter for setting the lower limit of oxygen for survival. It has already been shown in the case of goldfish (Fry and Hart, 1948), and for the speckled trout (Graham, 1949 and Job, 1955), that in the zone of respiratory dependence when the oxygen uptake is reduced to a level just equal to the standard rate, the animal has already reached the 'incipient lethal level' (Fry, 1947). At this level, the animal can just maintain itself and is capable of sustaining only the basic ancillary vital activities. But once the rate of uptake falls below this level, the animal can no longer tolerate oxygen lack and is bound, eventually to succumb; but how soon depends upon its ability to resist asphyxia. For a detailed analysis of the incipient lethal level, and its implications in the study of respiratory metabolism see Shepard (1955).



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