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EFFECTS OF THE P₀₂ ON SURFACING ACTIVITY AND FOOD UTILIZA-TION IN THE AIR-BREATHING PERCH ANABAS SCANDENS

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ABSTRACT

Fed Anabas scandens surfaced 555 times, swimming 278 m/day in non-aerated water (P_{0_2} : 66 mm Hg) and 855 times, travelling 428 m/day in aerated water (P_{0_2} : 147 mm Hg). Consumption, assimilation, production and metabolism of either series averaged 22, 18, 5 and 13 mg dry substance/g live fish-day, respectively. The high P_{0_2} elevated metabolism, increased surfacing and swimming activities but failed to alter food utilization. Culturing *A. scattdens* in aerated waters offers no special advantage. Starved fish in non-aerated and aerated waters surfaced 330 times and swam 164 m/day, expending 1.5 mg/g-day.

1. INTRODUCTION

Air-breathing fishes, inhabiting waters of low oxygen content in the tropics, surface intermittantly for "breathing air"; Pandian and Vivekanandan (1976) controlled the swimming distance of the obligatory air-breathing fish *Ophiocephalus striatus* by rearing them in cylindrical aquaria containing different depths of water. Since the fish were forced to swim to the surface in order to exchange atmospheric air, such an experimental design permitted the measurement of food intake, growth, and sustained active metabolism in relation to surfacing activity on a long term basis. The Authors found that the fish in deeper waters consumed significantly more food, swam longer distance and dissipated more energy on swimming and surfacing activities.

The P_{O_2} of several tropical ponds undergoes wide and rapid diurnal fluctuations, thereby exposing air-breathing fishes like *Anabas scandens* to a range of P_{O_2} from 46 mm Hg (at 6 a.m.) to 130 mm Hg (at 6 p.m.) in a day (e.g. Reddy, Natarajan 1970). The fluctuations in the P_{O_2} therefore may influence the frequency of surfacing (e.g. Hughes, Singh 1970b), which in turn, alters the energy required for such activities; i.e. the fluctuations in the P_{O_2} may ultimately influence the rate and efficiency of food utilization. The present paper reports on the effects of fluctuations in the P_{O_2} on surfacing activity and food utilization in the obligatory air-breathing fish *A. scandens*.

2. MATERIALS AND METHODS

A. scandens weighing 11 ± 2.2 g (mean \pm SD), 8 cm body length, was reared in cylindrical (15 cm diameter) aquarium (capacity: 3.75 l). A translucent glass plate of 14.5 cm breadth (30 cm length) was introduced into each aquarium: on one side of the glass partition, water was aerated; but the fish was confined to the other side. Water could move through the space available between the wall of the aquarium and the partition, while the fish swam in its semi-circular cylinder. This arrangement effectively minimized the horizontal swimming activity of the fish and also minimized the distraction of the fish from the bubbling stone connected to the aerator. The depth of water was kept constant at 25 cm in all aquaria and therefore the fish travelled 0.5 m when surfaced to breathe.

Two series were run; one in aerated aquaria and the other in non-aerated aquaria. In each series, one group was starved, and the other was fed goat liver twice every day for a period of two-

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hours. From control aquaria, fluids lost from the liver pieces during 2-hour period was measured and found to be $1.1\pm0.13\%$ of the liver. Unfed liver was collected from the experimental aquaria using a pipette and along with its respective weight, 1.1% was added as the correction for the lost fluids from the liver during the 2-hour feeding period. Faeces were collected by filtering the entire aquaria once every three days. The "Sacrifice Method" (Maynard, Loosli 1962) was used for determining the water content of the test individuals of both groups in each series before beginning the experiment.

The scheme of energy balance followed in the present work is that of IBP formula (Petrusewicz, Mac Fadyen 1970) usually represented as C = P + M + F, where C is the consumption of food, P—the production, M—the energy lost as heat due to metabolism and F—the faeces. The quantity of dry food assimilated (A) was estimated by subtracting the F from the C, and that of the P by subtracting the initial dry weight of the individual at the beginning of the experiment from the final dry weight of the individual at the end of the experiment. As the C, P and F are known, metabolism (M) can be calculated relating it to per unit initial live weight (g) of the fish per unit time (day). To express metabolism as ml O₂ uptake, 4.8 g cal energy was considered as equivalent to 1 ml of O₂ uptake (Engelmann 1966); calorific value of A. scandens was known to be 5000 g cal/g dry weight (see Golley 1961).

The experiment was conducted in a laboratory, where there was no disturbance except for feeding and routine observations; the laboratory was illuminated for 14 h/day from 6 a.m. to 8 p.m.; water temperature fluctuated between 25 and 27°C and averaged to 26°C. The number of times each test individual surfaced was observed for a known period of time (30 to 45 min), three times a day at 7 a.m., 1 p.m. and 7 p.m. The distance travelled per individual per day was estimated by multiplying the mean number of visits per unit observation time with twice the depth of water. Observations were made daily for a period of 28 days; since observations were made on three individuals in each of the groups belonging to two different series, each value in Tab. II represents the average (surfacing) performance of a minimum of 175 observations.

Following standard procedures, dissolved oxygen content (Winkler method) and free (dissolved) carbon dioxide content (Stroganov method) of each aquarium water were estimated every day at about 3 p.m. Following Pierce et al. (1973), the dissolved oxygen content values were converted to partial pressure of oxygen (P_{0_2}). For estimation of free carbon dioxide content, phenolphthalein was added as indicator to 100 ml of water sample. This was titrated against N/44 sodium hydroxide until pink colour disappeared. The amount of free CO₂ expressed in mg/l, was calculated multiplying the titre value by 10. Water was changed in the aquarium once every three days, except on the 24th day of the experiment, when the aquaria were left for five days without changing water.

3. RESULTS AND DISCUSSION

a. SURFACING AS FUNCTIONS OF FLUCTUATING PO2 AND CO2

Since water was changed in all the aquaria once every three days, the respective values obtained for the P_{O_2} and free CO_2 on the O day, the first and second days (refer Material and Methods) were separately averaged and are shown in Fig. 1. The significant drop (0 day as first day; Student's t=3.347; P<0.05) in the P_{O_2} of the non-aerated aquaria containing fed group may be due to decomposing faeces. On an average, the starved and fed groups in the aerated series enjoyed P_{O_2} of 152 and 147 mm Hg, and CO_2 of 1.5 and 3.5 mg/l, while those of the non-aerated series were exposed to the P_{O_2} of 111 and 66 mm Hg, and CO_2 of 3.8 and 5.4 mg/l, respectively.

Although both the fed and starved groups in the aerated series enjoyed an average P_{O_2} of about 150 mm Hg, they still surfaced once every 1.7 and 4.2 min, respectively; this clearly shows that the gills are inadequate to supply the oxygen requirements entirely from water. Under similar situations (25°C; 148 mm Hg P_{O_2}), *A. testudineus* (22 g) is reported to surface once every 12.2 min (Hughes, Singh 1970b). The fact that the proportion of oxygen consumption obtained from air is only 54% in *A. testudineus* (Hughes, Singh 1970a) and 80% in *A. scandens*

Effects of the Po, in A. scandens



Fig. 1. Fluctuations in the P_{02} (A) and free CO₂ (B) in the aerated (1 and 3) and non-aerated (2 and 4) waters of aquaria containing fed (3 and 4) and starved (1 and 2) *Anabas scandens*. The values for the 0, first and second days represent the average of minimum 3 values estimated on 8 different dates; third, fourth and fifth days represent the 26th, 27th and 28th days of the experiment

(Reddy, Natarajan 1970), explains at least in part why the latter surfaces more frequently than the former.

The feeding group of the aerated series surfaced around 850 times/day, despite the variations in the P_0 from 155 mm Hg on the 0 day to 107 mm Hg on the 4th day (Fig. 2A). The differences observed in the number of surfacings among the starved groups of either series were not significant, (Student's t=5.780; P<0.10) and the surfacing, which was dependent on the Po, decreased from 377 times/day at 155 mm Hg to 253 times/day at 83 mm Hg. In the fed group of the non-aerated series, the surfacing increased from 501 times/day at 134 mm Hg on the O day to 651 times/day at 22 mm Hg on the 3rd day (Tab. I) and subsequently decreased (Fig. 2A). The analyses on the number of surfacings as function of the fluctuating Po, on different days of the experiment (Fig. 3) reveal that when the Po, decreased below 60 mm Hg on different days of the experiment, the fish increased the number of surfacings from about 550 to 650 times/day and perhaps obtained less proportion of oxygen via gills. Hill et al. (1972) also noted that the young dipnoan fish Lepisosteus oculatus increased surfacing from about 190 times/day at the Po, of 80 mm Hg to over 480 times/day, when the Po, decreased below 79 mm Hg (however, see also De Roth (1973). The bowfin Amia calva increased surfacing to over 1,200 times/day below 100 mm Hg at 30°C, over 440 times/day below 80 mm Hg at 20°C and over 190 times/day below 40 mm Hg at 10°C (Johansen 1970). Vivekanandan (1975) observed that the obligatory air-breathing fish O. striatus increased

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Fig. 2. Effects of the P_{O_2} (A) and free CO₂ (B) on the number of surfacings by *Anabas scandens* 1-4 as in Fig. 1. The values for the 0, first and second days represent the average performance of minimum 3 individuals observed on 8 different dates for a period of about 3 hours per day

Table I. Effect of P_{0_2} and free CO₂ on the surfacing activity of fed *Anabas scandens* of the nonaerated series and correlation coefficients *r* between them. Surfacing values represent the average performance of 3 individuals (mean ±SD), observed for a period of 28 days at 26°C

Number of days after aquarium water change	Po	Free CO ₂ (mg/l)	Surfacing (times/day)	Coefficients of correlation		
	(mm Hg)			Surfacing on Po ₂	Surfacing on CO ₂	
0	134±18.3	1.9 ± 0.01	501 ± 20.8	-0.84**	0.41*	
1	40 ± 10.0	5.6 ± 0.72	570 ± 61.6	-0.96***	0.72**	
2	22 ± 0.00	8.6 ± 0.65	633 ± 30.6	-0.94***	0.79**	
- 3	22 ± 0.00	9.1±0.00	651 ± 60.0	-1.00***	1.00***	

* Not significant P>0.1. ** Significant P<0.02. *** Highly significant P<0.001.

the number of surfacings from 912 times/day at 155 mm Hg to 1183 times/day at the P_{O_2} of 42 mm Hg. The facultative air-breathing catfish *Heteropneustes fossilis* increased surfacing, when the P_{O_2} decreased below 70 mm Hg (Arunachalam et al. 1976). Feeding *A. scandens* of the aerated series undertook more or less constant number of surfacings (850 times/day), though the CO₂ significantly increased from 1.2 mg/l on the 0 day to 8.6 mg/l on the 5th day (Fig. 2B). Surfacing response of the feeding group of the non-aerated series differed from that of the aerated series; it increased from about 501 times/day at 1.9 mg CO₂/l to 651 times/day at 9.1 mg CO₂/l (Tab. I). This observation is again supported by the analyses of the number of surfacings as function of the fluctuating CO₂ on different days of the experiment (Fig. 3). When the CO₂ increased above 5.4 mg/l on different days, the



Fig. 3. Day-to-day fluctuations in the P_{0_2} (A) and free CO₂ (B) in the non-aerated aquaria and the consequent differences in the surfacing response (C) of the fed *Anabas scandens*. Each surfacing value represents the average performance of minimum 3 individuals observed for about 3 hours per day for a total period of 28 days

fish surfaced more frequently and perhaps obtained less oxygen branchially. On the whole, the surfacing response of the fed group in the non-aerated series increased until the P_{O_2} of 22 mm Hg and CO₂ of 9.1 mg/l, which were the levels usually observed on the 3rd day in the aquaria.

The starved groups of either series decreased surfacing from about 380 times/day at 1.2 mg CO_2/l to 260 times/day at 5.9 mg/l. To both the increasing CO_2 and decreasing Po, levels, the level and pattern of response of the starved groups of either series were different from those observed for the regularly fed groups. Arunachalam et al. (1976) also observed that the surfacing response of the starved H. fossilis was different from that of regularly fed animals. They reported that the number of surfacings linearly decreased from about 180 times/day at the Po, of 140 mm Hg to about 150 times/day at 115 mm Hg. In order to assess the relative importance of the Po, and CO2 on air-breathing, correlation coefficients for surfacing-Po, and surfacing-CO₂ were calculated for the fed group of the non-aerated series (Tab. I); the coefficients obtained for surfacing-Po, are highly significant on 1st, 2nd and 3rd day after the change of water in aquaria, while those obtained for the surfacing- CO_2 are only significant on the 1st and 2nd days and high significant on the 3rd day. Therefore, the surfacing activity is primarily dependent on the Po, between 40 and 22 mm Hg. Within the range of CO₂ from 5.6 to 8.6 mg/l, the surfacing activity may be of secondary importance. As the coefficient became highly significant for the surfacing-CO₂ (at 9.1 mg free CO₂/l) on the 3rd day, breathing activity may be dependent on the levels of free CO₂ as well as Po. Hughes and Singh (1970b) concluded that total CO_2 is the most important factor in determining aquatic and/or aerial respiration in A. testudineus; the climbing perch performed aquatic and aerial respiration upto the Po, of about 19 mm Hg in water, provided the total CO2 content of the water was below 15 vols % and at 16 vols %, aerial respiration became predominant and exclusive between 18 and 22 vols %. In the present study, A. scandens was subjected to maximum free CO₂ stress of less than 9.9 mg/l, which may be approximately equivalent to the total CO₂ content of less than 10 vols %, and hence the results obtained and the conclusions drawn in the present study may not be comparable to those of Hughes and Singh (1970a, b). Whereas the lowest Po, to which A. testudineus exposed by Hughes and Singh (1970b) was 18.9 mm Hg, that to which the fed A. scandens of the non-aerated series subjected was as low as 18 and 13 mm Hg on the 4th and 5th day following water change in aquarium. Between 13 and 22 mm Hg, A. scandens continued surfacing but reduced the number of times it visited the surface (Fig. 2A). The fact that A. scandens reduced the number of surfacings at such low Po, suggests that the decreasing Po, reduced the metabolic level, which in turn, decreased the number of visits to the surface forcing the fish to obtain proportionately less oxygen from the air.

b. SURFACING AND DISTANCE TRAVELLED

Hitherto analyses were made on the data obtained for the P_{O_2} and free CO_2 and the consequent surfacing response of the different groups. However, certain other trends became apparent, when the mean data (Fig. 4) obtained for the num-

ber of surfacings and distance travelled/day for the total period of 28 days were averaged. The fed group in the non-aerated series, which was subjected to an average P_{O_2} of 66 mm Hg, surfaced 555 times and swam a distance of 278 m/day. The number of surfacings as well as the distance travelled by the fed fish of the aerated series, enjoying the mean P_{O_2} of 147 mm Hg, significantly increased to 855 times and 428 m/day (Tab. II). The strong positive correlation observed between the mean P_{O_2} and the number of surfacings is both interesting and unexpected, for it is commonly believed that the surfacing activity of the air-breathing fish decreases with the increasing P_{O_2} (see Johansen 1970). Recalculation of the data reported by Hughes



Fig. 4. Effects of aeration on the number of surfacings and distance swum by the fed (1) and starved
 (2) Anabas scandens reared in aquaria containing aerated (A) and non-aerated (B) waters at 26°C.
 Each value represents the average performance of minimum 3 individuals (mean ±SD) observed for 30 to 45 minutes, 3 times a day for a period of 28 days

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and Singh (1970b) reveals that A. testudineus (22 g) also increased the number of surfacings from 90 times/day at the P_{O_2} of 87 mm Hg to 105 and 120 times/day with increasing P_{O_2} of 99 and 150 mm Hg, respectively. An environmental factor like the P_{O_2} is suggested to primarily influence the metabolism of the fish, perhaps at cellular level, and its effect on activity may be secondary (see Shelton 1970). The high P_{O_2} to which A. scandens and A. testudineus were exposed appears therefore to have first elevated the metabolic level, which in turn, increased the number of surfacings to procure proportionately more oxygen via the air-breathing organs.

The surfacing response of the starved A. scandens to the decreasing P_{O_2} was different. Starved groups exposed to an average P_{O_2} of 152 (aerated water) and 111 (non-aerated water) mm Hg surfaced about 330 times, travelling 165 m/day (Tab. II). The stress of decrease in the P_{O_2} in the non-aerated series was perhaps not sufficient to produce any marked effect on surfacing activity. Starved *H. fossilis* exposed to an environmental stress of different aquarium depths exhibited no difference in swimming activity up to a distance of 0.8 m/surfacing and reduced the surfacing, when it had to swim more than 0.8 m/surfacing (Arunachalam et al. 1976).

c. FOOD UTILIZATION AS FUNCTION OF THE PO2

Continuous aeration did not influence food consumption significantly, as it was about 22 mg/g-day for the aerated and non-aerated series. The differences observed between the rates of assimilation (18 mg/g·day) and production (5 mg/g· day) among the aerated and non-aerated series were also not statistically significant (Tab. II). The minor difference observed in the consumption of food among the series became insignificant due to clear difference observed in the assimilation efficiency. While the group in the non-aerated aquaria assimilated 90% of the food consumed, that in the aerated aquaria assimilated the food with 82% efficiency. Assimilation efficiency is known to average around 80% in several fish species (Winberg 1956), and it did not vary significantly as functions of body size (Gerking 1952, 1971; Pandian 1967a, b) and nutritional state (Pandian 1967d) of the fish as well as temperature (Davies 1964; Vivekanandan 1975), quality (Pandian 1967b) and quantity (Gerking 1955, 1971; Pandian 1967c; Vivekanandan 1976) of food organisms. Net production efficiency (K_2) was also about 27% in either series and did not differ markedly. It may be concluded that while the increase in the Po, increased surfacing activity, its influence on the rate and efficiency of food utilization is not significant. Therefore culturing A. scandens in aerated waters may have no advantage over culturing in non-aerated water.

d. LEVELS OF METABOLISM

While energy expended on the metabolic processes by the feeding groups was equivalent to about 13.4 mg dry fish substance/g live fish day (=0.58 ml O₂ upta-ke/g·h), that expended by the starved group in either series amounted to 1.5 mg/g·day (=0.065 ml O₂ uptake/g·h) (Tab. II). The total O₂ uptake of *A. scandens* is reported to range from 0.21 to 0.26 ml/g·h (Reddy, Natarajan 1970, 1971) and it is 0.11 ml/g·h for *A. testudineus* (Hughes, Singh 1970a). *A. testudineus* was fed

Table II. Effects of the P_{O_2} on surfacing activity and food utilization in *Anabas scandens*. Each value represents the average performance of 3 individuals (mean \pm SD) observed for a period of 28 days at 26°C; Students "t" test has been used to determine the levels of significance

	Aerated		Non-aerated	
Parameters	Fed (Po ₂ 147 mm Hg)	Starved (Po ₂ 152 mm Hg)	Fed (Po ₂ 66 mmHg)	Starved (Po2 111 mm Hg)
Number of surfacings/day	855 ± 92.4^{a}	344±25.6 ^b	555 ± 43.5^{a}	311±20.0 ^b
Distance travelled (m/day)	428 ± 46.2	172 ± 12.8	278 ± 21.8	156 ± 10.0
Consumption of food (C) $(mg/g \cdot day)$	$23.8 \pm 2.02^{\circ}$		$19.3 \pm 0.85^{\circ}$	-
Assimilation (A) $(mg/g \cdot day)$	19.4 ± 1.86^{d}		17.6 ± 0.72^{d}	·
Production (P) $(mg/g \cdot day)$	$+5.1\pm0.91$	-1.5 ± 0.17	$+5.1 \pm 0.30^{e}$	-1.5 ± 0.20
Metabolism (M) (mg/g·day)	$14.3 \pm 2.14^{\circ}$	1.5 ± 0.17	12.5 ± 0.32	1.5 ± 0.20
Metabolism (M) (ml $O_2/g \cdot h$)	0.62	0.065	0.54	0.065
Assimilation efficiency (A/C)	81.9		89.5	
Gross production efficiency $(K_1 = P/C)$ (%)	21.6		25.9	
Net production efficiency $(K_2 = P/A)$ (%)	26.4		28.9	-

a. Significant; t=4.186; p **≪0.02**

c. not significant; t=1.906; p ≪0.10

e. not significant; 1=0.770; p ≤0.10.

b. not significant; t=5.780; p < 0.10d. not significant; t=1.282; p < 0.10 by Hughes and Singh (see 1970b), once in 3 or 4 days and they offered no food 1 or 2 days prior to O_2 uptake estimates; Reddy and Natarajan (1970, 1971) gave no food to *A. scandens* on the experimental day. Therefore, the O_2 uptake values reported by Hughes and Singh (1970a), Reddy and Natarajan (1970, 1971) may at best be compared with those obtained for the starved *A. scandens*. This aspect has been dealt elsewhere (Pandian, Vivekanandan 1976; Arunachalam et al. 1976).

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4. SUMMARY

Reared in cylindrical aquaria, fed Anabas scandens surfaced 555 times, travelling 278 m/day in non-aerated water (mean P_{O_2} :66 mm Hg) and 855 times swimming 428 m/day in aerated water (147 mm Hg). Food consumption, assimilation, production and metabolism of both series averaged to 22, 18, 5 and 13 mg dry substance/g live fish-day, respectively. The high P_{O_2} elevated metabolism, which in turn increased surfacing and swimming activities but it did not alter the pattern of food utilization. Culturing A. scandens in aerated water offers no advantage over non-aerated water. Since water was changed once every 3 days, the P_{O_2} in the non-aerated aquarium of the fed group decreased from 134 to 42 and 23 mm Hg on the 1st and 2nd day. Analyses of day-to-day fluctuations in the P_{O_2} and free CO_2 as function of surfacing reveal that A. scandens increased surfacing from 550 to 650 times/day below the P_{O_2} of 60 mm Hg and above 5.4 mg CO₂/l; surfacing is dependent more on the P_{O_2} than on CO_2 . Starved groups in aerated and non-aerated aquaria surfaced 330 times and swam 164 m/day, expending 1.5 mg/g.day.

5. PE3HOME

Выращиванные в цилидрических аквариумах, кормленные A. scandens в неаэрированной воде (в среднем Ро.: 66 мм рт. ст.) всплывали на поверхность 555 раз в сутки, совершая таким образом путь длиной в 278 м. В аквариумах с аэрированной водой (147 мм рт. ст.) они всплывали 855 раз в сутки, делая при том 428 м. Потребление пищи, ассимиляция, продукция и траты на обмен составляли соответственно 22, 18, 5 и 13 мг сухого веса/г сырого веса рыбы в сутки. Высокое Род повышало траты на обмен, что в свою очередь увеличивало частоту всплывания и повышало двигательную активность, но не изменяло образа использования пищи. Выращивание A. scandens в аэрированной воде не имеет никаких преимуществ по сравнению с вырашиванием их в неаэрированной воде. Воду изменяли каждые 3 дня, вследтвие чего в аквариуме с аэрированной водой, в котором находились кормленные A. scandens, Po2 снижалось с 134 до 43 и 23 мм рт. ст. на первый и второй день. Анализ изменений Роз и свободного CO2 как функция частоты всплывания показывает, что A. scandens учащает это действие с 550 до 650 раз в сутки при Ро2 ниже 60 мм рт. ст. и CO2 выше 5.4 мг/л; частота всплывания зависит в большей степени от Ро2, чем от СО2. Некормленные группы A. scandens в обсих сериях (в аэрированной и неаэрированной воде) всплывали 330 раз, делая 164 м в сутки; траты на обмен составляли 1.5 мг/г в сутки.

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