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IMPORTANCE OF AIR AND WATER BREATHING IN RELATION TO SIZE OF THE AFRICAN LUNGFISH *PROTOPTERUS AMPHIBIUS* PETERS*

BY K. JOHANSEN AND J. P. LOMHOLT
Department of Zoophysiology, Aarhus University, DK-8000 Aarhus, Denmark

AND G. M. O. MALOIY
Department of Animal Physiology, University of Nairobi, Nairobi, Kenya

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SUMMARY

1. Oxygen uptakes from air and water have been measured in relation to weight of the African lungfish *Protopterus amphibius* Peters.
2. Combined O_2 uptake from air and water ranged from 60 ml O_2 kg^{-1} h^{-1} STPD, in a 3·7 g specimen, to 30 ml O_2 kg^{-1} h^{-1}, in a 255 g specimen.
3. While the combined O_2 uptake changed by a factor of 2, within the weight range under study, the aquatic O_2 uptake changed 8-fold within the same range. The smaller fish satisfy 70% of their O_2 requirement by aquatic breathing compared to 10–15% in the grown specimens.
4. The pattern of bimodal breathing in *P. amphibius* is discussed in relation to the natural habitat of the species.

INTRODUCTION

All air breathing fishes are bimodal breathers and depend to a variable extent on water and air breathing to satisfy their metabolic requirements. In some species (e.g. the bowfin, *Amia calva*) the relative importance of air and water breathing is highly temperature dependent. A change in temperature from 10 to 27 °C will alter O_2 absorption from nearly complete to about 30% aquatic (Johansen, Lenfant & Hanson, 1970). The adult African and South-American lungfishes are obligate air breathers and the temperatures of their natural habitats change little. The African lungfish, *Protopterus aethiopicus*, relies on air breathing for about 90% of its requirement at 25 °C (Lenfant & Johansen, 1968). Among other air breathing fishes the Australian lungfish, when at rest in normal conditions, is purely aquatic and the gills suffice for absorption of its entire O_2 requirement (Lenfant, Johansen & Grigg, 1966). During increased physical activity the lung becomes functional as an accessory O_2 absorber. The adult South-American lungfish, *Lepidosiren*, cannot

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dispense with air breathing. Thus Sawaya (1946) reported that adults depend on air breathing for 96% of their O_2 requirement, at 20 °C, although in juvenile individuals aquatic O_2 uptake exceeds aerial absorption at 18 °C (Johansen & Lenfant, 1967). No systematic study appears, however, to have been made of the relative importance of aquatic and aerial breathing during development and growth of any species of lungfish, although McMahon (1970) stated that dependence on aerial respiration is less developed in juvenile specimens of *Protopterus*.

Hughes *et al.* (1973, 1974) have traced a weight specific increase in the surface area of air breathing organs of the teleosts *Anabas testudineus* and *Amphipnous cuchia* during growth. This information is suggestive that the relative importance of aerial and aquatic breathing may change during growth in bimodal breathers among teleost fishes.

The eggs of lungfishes develop in water and the parental care includes a stirring or agitation of the water by the male (Johnels & Svensson, 1954). This activity is most certainly of importance for the O_2 supply to the developing eggs or newly hatched larvae. It has been observed that young of *P. annectens* start gulping for air at the surface, at lengths of 25 mm, prior to the beginning of active feeding, which does not start until the yolk sac is consumed at a length of 55 mm (Johnels & Svensson, 1954). Smith (1931) observed larvae of *P. aethiopicus* of about 3 cm in length, to gulp air at the surface, but only at very rare intervals.

The objective of the present study has been to measure the extent of aquatic and aerial O_2 uptake in relation to the size of *P. amphibius*. The smallest specimen used weighed about 4 g, while the largest were adults of about 500 g.

MATERIAL AND METHODS

The African lungfish *Protopterus amphibius* Peters is distributed in north-eastern Kenya primarily in the drainage area of Tana river. The specimens used in the present study were collected in a lake near Malindi, Kenya. The smaller specimens were collected in the aestivating state and brought back to a non-aestivating condition by return to water. The lungfishes were air-freighted to Aarhus, Denmark, where they were kept for 1 year in aquaria at 30 °C prior to this study. The animals were fed on beef twice weekly.

Oxygen uptake from air and water of the smaller specimens were measured at 30 °C using a technique slightly modified from that described by Emilio & Shelton (1974). A plexiglass container was filled with aerated water to a level as indicated in Fig. 1. The amount of O_2 absorbed by air breathing from the enclosed gas space in the cylinder was measured by periodically analysing the gas composition of the air phase. This was done by displacing the gas, using syringe D, into a Radiometer cuvette for measurement of P_{O_2} with an O_2 electrode (Radiometer). After a reading new air was drawn into the cylinder (stopcock A). During a breathing period the pressure in the system was maintained at the same level by connecting the enclosed air space to the glass tube (B) which had a movable fluid meniscus. O_2 uptake from water by gill and skin breathing was measured by periodic analysis of the water P_{O_2}, drawing water samples from stopcock C. The oxygen electrode was calibrated with air and air-equilibrated water, depending on the sample to be analysed.

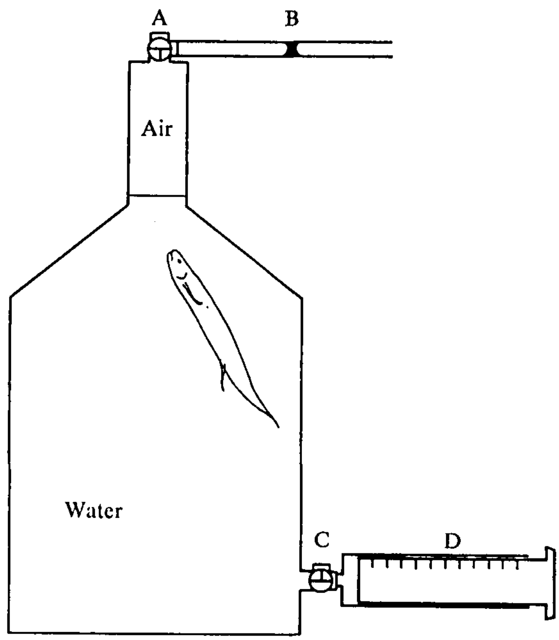

Fig. 1. Respirometer used for O_2 uptake measurements from air and water in small specimens of *P. amphibius*.

The area of the air/water interface was relatively small and the timing of sampling was arranged so as to minimize air to water P_{O_2} gradients. In this way gaseous exchange between the media was kept small enough to be neglected. O_2 uptake of the larger fishes was measured in a respirometer with the gas phase connected to a differential manometer. CO_2 being absorbed from the gas phase by 'carbosorb' (BDH-chemicals) the aerial O_2 uptake was measured as the amount of pure O_2 which had to be injected into the gas phase to restore the manometer reading to zero from time to time. Water O_2 uptake was measured by analysis of water samples as described above. Although the interface between water and gas was larger in the latter apparatus, blind checks showed that gas exchange across the surface was so small as to be insignificant. All O_2 uptakes are expressed as volumes at STPD conditions.

RESULTS AND DISCUSSION

The combined O_2 uptake from air and water ranges from 60 ml O_2 STPD kg^{-1} h^{-1} in the smallest (3·7 g) fish to about 30 ml O_2 kg^{-1} h^{-1} in one of the larger fish (255 g) (Fig. 2). The large variability of individual measurements on each fish may have been caused by differences in activity and feeding schedule. Smith (1935) reported a marked influence on O_2 uptake related to feeding and starvation in *P. aethiopicus*. Whereas the overall O_2 uptake changed by a factor of 2 (comparing the smallest to the largest fishes) the oxygen uptake from water changed 8-fold, being reduced from 40 ml O_2 kg^{-1} h^{-1} in the smallest to 5 ml O_2 kg^{-1} h^{-1} in the largest specimens. Evaluated on a per cent basis (Fig. 2, bottom) the results demonstrate a marked change in the pattern of bimodal gas exchange in *P. amphibius*.

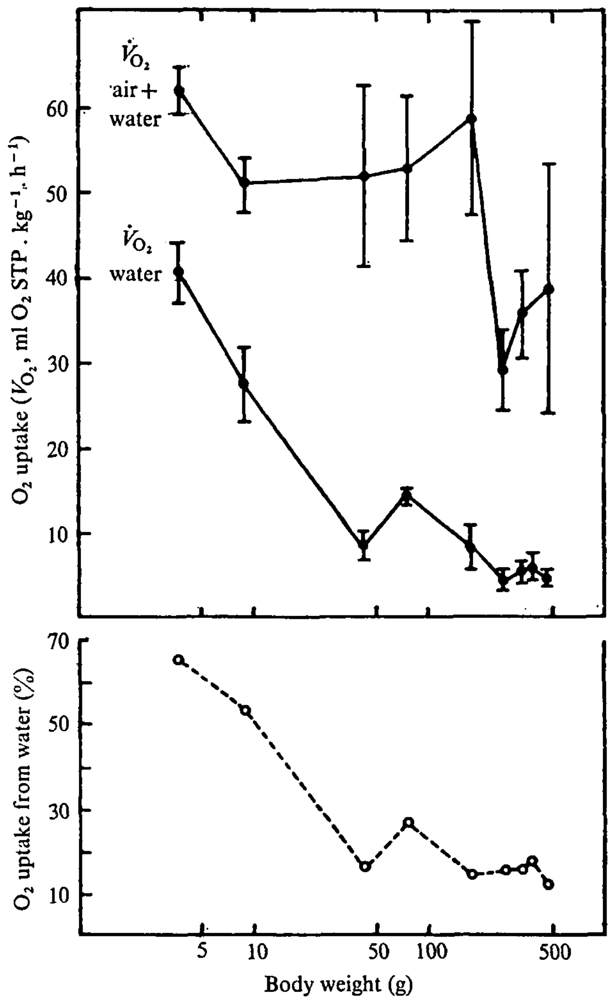

Fig. 2. Oxygen uptake related to size of *P. amphibius*. Top curve shows combined O_2 uptake from air and water. Middle curve shows O_2 uptake from water. At bottom per cent O_2 uptake from water is depicted. 3–7 experimental runs were done on each fish. Results presented as mean ± S.D.

While fish in the immediate post larval and young stages depend on water breathing for 70% or more of their oxygen requirement, grown fish between 300 and 500 g obtain as little as 10–15% of their oxygen from the water.

The O_2 content of the water in the respirometers was never reduced to less than two thirds of aeration value during the course of an experiment. In its natural habitat grown specimens of *P. amphibius* were, however, often found in very O_2 deficient water (i.e. $P_{I_{O_2}}$ less than 30 mmHg) suggesting that near total dependence on air breathing may prevail under natural conditions. Small and young fish were never collected in severely O_2 deficient water and when placed in O_2 deficient water in aquaria the small fish stayed close to the surface in better aerated water. Present observations also suggest that the young fish increase the frequency of air breathing in O_2 deficient water while an earlier study revealed that the air breathing pattern of grown specimens of *P. aethiopicus* was unaffected by the level of oxygenation in the water (Johansen & Lenfant, 1968).

The present results suggest that the African lungfish, *P. amphibius*, only gradually develops the faculty of extracting the total or nearly all of its oxygen requirement from atmospheric air. Moreover, *P. amphibius* among all species of *Protopterus* retains external gills and large-brimmed, well-vascularized anterior fins also in the adult stage. The altered dependence on water and air breathing during the life cycle of *P. amphibius* will most likely set territorial limits to the distribution of larvae and young fish within the distribution range of the species.

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