

AIR-BREATHING DURING ACTIVITY IN THE FISHES *AMIA CALVA* AND *LEPISOSTEUS OCULATUS*

C. G. FARMER* AND D. C. JACKSON

Department of Molecular Pharmacology, Physiology and Biotechnology, Brown University, Providence, RI 02912, USA

*Present address: Department of Ecology and Evolutionary Biology, University of California at Irvine, Irvine, CA 92717, USA
(e-mail: cfarmer@uci.edu)

Accepted 19 January; published on WWW 5 March 1998

Summary

Many osteichthyan fishes obtain oxygen from both air, using a lung, and water, using gills. Although it is commonly thought that fishes air-breathe to survive hypoxic aquatic habitats, other reasons may be more important in many species. This study was undertaken to determine the significance of air-breathing in two fish species while exercising in oxygen-rich water. Oxygen consumption from air and water was measured during mild activity in bowfin (*Amia calva*) and spotted gar (*Lepisosteus oculatus*) by sealing a fish in an acrylic flume

that contained an air-hole. At 19–23 °C, the rate of oxygen consumption from air in both species was modest at rest. During low-level exercise, more than 50 % of the oxygen consumed by both species was from the air (53.0±22.9 % *L. oculatus*; 66.4±8.3 % *A. calva*).

Key words: bowfin, *Amia calva*, gar, *Lepisosteus oculatus*, air-breathing, activity, myocardial oxygenation, evolution, lungs, Osteichthyes.

Introduction

An internal sac that functions to absorb atmospheric oxygen is found in many vertebrates (Fig. 1). In basal fishes and in tetrapods, this organ is commonly called a lung. Mapping the presence of lungs onto a vertebrate phylogeny suggests that lungs are primitive structures that precede in evolutionary time the swimbladders of teleosts, which function primarily in buoyancy and sound transmission (Brainerd, 1994; Liem, 1988). Because lungs are found in the Tetrapoda, Dipnoi (lungfishes), Polypteridae (the bichirs), Lepisosteidae (gars) and *Amia* (bowfin), they were probably present in the common ancestor of the Actinopterygii and Sarcopterygii, thus dating the evolution of lungs to at least as early as the Late Silurian (over 400 million years ago).

The presence of a lung in an animal was at one time considered a feature that distinguished tetrapods from fishes (Gunther, 1871). When it was discovered that fishes also have lungs, their function was tied to a hypoxic habitat. This is exemplified by early writings on the discovery of the Australian lungfish: 'When we recollect that the animal evidently lives in mud or in water charged with the gases which are the product of decomposing organic matter, the usefulness or necessity of such an air-breathing apparatus, additional to the gills, becomes at once apparent' (Gunther, 1871). Since the establishment of this idea, data have emerged suggesting that air-breathing is not necessarily correlated with a hypoxic habitat (Kramer *et al.* 1978; Rahn *et al.* 1971). For example,

measurements of oxygen content throughout the habitat of the Australian lungfish revealed that it is not hypoxic, even during periods of drought (Grigg, 1965). Rather than using lungs to survive hypoxic water, these animals use lungs to support activity (Grigg, 1965).

Air-breathing has been linked to activity in many other primitive fishes: *Polypterus senegalus*, *Amia calva*, *Lepisosteus oculatus* and *Megalops atlanticus*, a primitive teleost, (Becker, 1983; Johansen *et al.* 1970; Magid, 1966; Reighard, 1903; Saksena, 1963; Shipman, 1989; Schlaifer and Breder, 1940). The present study was undertaken to quantify the amount of oxygen consumed from the air and from the water by *A. calva* and *L. oculatus* while active in oxygen-rich water.

Materials and methods

Animals

Amia calva Linnaeus (bowfin) and *Lepisosteus oculatus* (Winchell) (spotted gar) were collected by electroshocking from the wild in April, in conjunction with the Florida Freshwater Fish and Game Commission and the National Biological Survey. They were held for 1 week in Florida and then transported by land to Rhode Island in 1211 (32 gallon) plastic containers with 0.05 % salt water and a mild dose of sedative (Trance; half to a quarter of the dose recommended

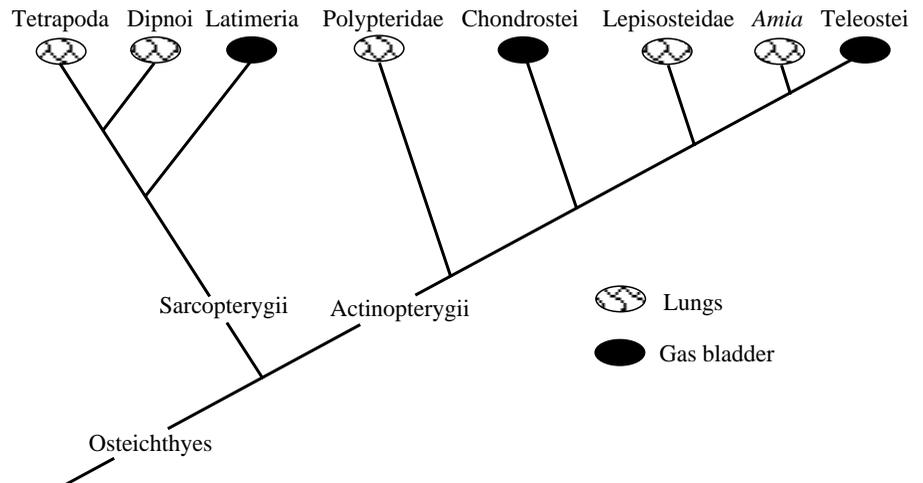


Fig. 1. Phylogenetic hypothesis of the relationships between extant fishes showing the presence or absence of lungs and a gas bladder (phylogeny based on Lauder and Liem, 1983).

by the manufacturer, Argent). The animals were housed indoors at 24 °C in 9461 (250 gallon) tanks with recirculating, buffered (pH 7.5) water. The *A. calva* were separated to prevent fighting, while the *L. oculatus* were allowed access to all areas of the tanks. They were fed a diet of live goldfish, frozen smelts, frozen catfish, live earthworms and trout pellets. They were exposed to plant growth lights and fluorescent lights for a photoperiod of 14 h:10 h light:dark. Initial infestations of *Argulus*, a parasitic decapod, were treated with trichlorfon (5 mg l⁻¹). Secondary bacterial infections were treated with the antibiotic oxytetracycline (2.64 mg l⁻¹). The *A. calva* weighed approximately 1 kg and were approximately 30 cm long. The *L. oculatus* weighed approximately 300 g and were approximately 25 cm long.

Training

An animal was netted from the holding tank and placed in a flume (covered with a black cloth) that contained water from the holding tank. Ammonia levels were monitored regularly and were maintained below 0.01 p.p.m. The animal was left undisturbed for up to a week to acclimate to the new environment. Training began with swimming periods of approximately 20 min at slow speeds (0.5 m s⁻¹). As the fish became accustomed to swimming in the flume, the duration of the training session was increased. The fish were not adept at finding the air-hole at higher speeds. They seemed confused and attempted to surface at other places in the flume. Hence, the swimming speeds were kept low so that the fish could easily find the air-box when desired. If the fish refused to swim and rested against the back of the flume, a mild electrical shock could be used to induce swimming.

Flumes

Any method that stimulated movement in the fishes would allow oxygen consumption measurements at rest to be compared with activity. The experiments carried out by Grigg (1965) entailed placing a fish in a tank with a stir bar. The movement of the bar and the water caused the fish to swim. Other means could have been used to stimulate movement, e.g.

chasing the fish with a stick. Although a flume was used to stimulate activity, these studies were not locomotor studies and the fluid dynamics of the experimental apparatus was not a major concern. Furthermore, there was no attempt to find the maximum sustainable speeds of the fish (U_{crit}). The fish were swimming relatively slowly and could sustain this movement continuously for more than 2 h.

Two flumes were constructed out of 1.3 cm (1/2 inch) Plexiglas. One was used for the experiments on *A. calva* (Fig. 2) and was rectangular with inside dimensions of 213.4 cm × 50.8 cm × 25.4 cm and contained 275 l of water. A central Plexiglas divider 1.3 cm thick separated the channels. Concentric, semi-cylindrical plastic wave guides directed the water around the ends of the central divider. A plastic grid was glued into one of the channels to create a swimming chamber 121.9 cm × 25.4 cm × 25.4 cm. At the back of the swimming channel, a metal grid was connected to a variable d.c. power supply that could be used to induce swimming. Over the swimming chamber, the lid was cut to form an air-breathing hole and a triangular covering of plexiglas was sealed over this

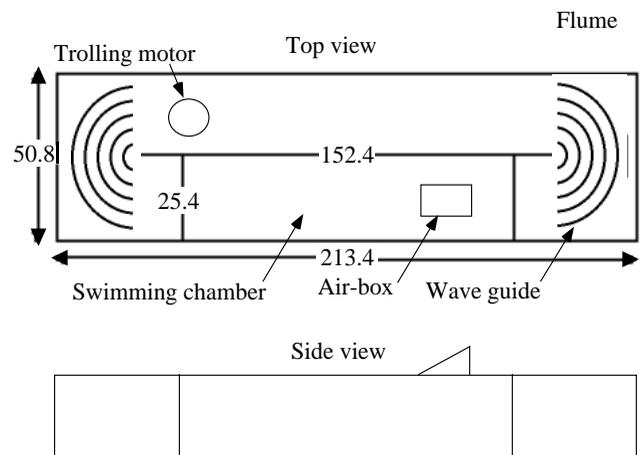


Fig. 2. Schematic diagram of the flume used to measure oxygen consumption from air and water in *Amia calva*. Dimensions are in cm.

hole. The air-box dimensions were 20.3 cm×15.2 cm×5.1 cm. Rubber weather-stripping was glued to the rim of the flume and brass buckles, attached with screws to the Plexiglas, were used to pull the lid tight against the weather-stripping. This construction produced a sealed system in which the circulating water was exposed to air only in the relatively small area of the breathing hole.

One end of the flume was raised 1.3 cm above the other so that gas bubbles that escaped from the air-hole would migrate to points where they could be collected through aquarium valves into glass syringes. A battery-powered trolling motor was used to move the water. The water speeds achievable with this motor ranged from 0.5 to 1.1 m s⁻¹. Water speed was measured in the middle of the swimming chamber using an electromagnetic flow meter (model 2000; Marsh-McBirney). There were no stagnant regions where the fish could remain stationary.

A second flume was constructed for *L. oculatus*. The design was the same as above, except that the inside dimensions of the flume were 121.9 cm×30.5 cm×15.2 cm and the flume held a volume of 56.6 l. The air-box was 20.3 cm×14.6 cm×5.1 cm. Additionally, a smaller trolling motor was used.

Oxygen consumption from water

To measure oxygen consumption from water, a peristaltic pump (Sigmamotor T8) continuously pulled water from the flume through tubing (Tygon) into a Plexiglas holder that contained an oxygen electrode (YSI electrode connected to an OM 200 meter). The water was returned to the opposite side of the flume. Humidified nitrogen- and air-saturated water were used to calibrate the electrode, and periodic checks of the calibration were performed during the experiment.

Oxygen consumption from air

Air was pulled by a pump (Applied Electrochemistry; R2 flow control unit) through the air-box into a glass tube containing Drierite (anhydrous calcium sulfate) and then through an oxygen analyzer (N-22M; Applied Electrochemistry). The rate of flow of air through the box was measured using a precision bore flowmeter (Lab Crest Division). Both *A. calva* and *L. oculatus* tend to exhale as they break the surface of the water (Deyst and Liem, 1985; Rahn *et al.* 1971). This exhalation into the air-hole diluted the oxygen flowing through the box, which caused a decrease in the percentage of oxygen flowing through the analyzer. The Fick principle was used to calculate the oxygen consumed from the air; this equation is as follows:

$$\dot{V}_{O_2} = ([O_2]_{in} - [O_2]_{out}) \times \text{airflow},$$

where \dot{V}_{O_2} is the rate of oxygen consumption from the air by the fish, $[O_2]_{in}$ is the concentration of oxygen of the air flowing into the box, $[O_2]_{out}$ is the concentration of oxygen of the air flowing out of the box and airflow is the flow of air through the box.

Gases that escaped into the flume from the air-hole were collected through aquarium valves by syringe and injected back into the air-hole.

Experimental protocol

The terms activity, exercise, movement, swimming, etc., are used throughout this paper to indicate a fish that is not staying still. They are not used in a quantitative manner (i.e. exercise does not imply a greater or lesser degree of movement than activity, etc.).

Preliminary experiments were carried out to determine whether sufficient rates of air-breathing occurred during activity to warrant construction of the flumes and further investigation. Six *A. calva* were monitored in an open flume. A fish was observed at rest for at least 1 h, then the fish swam for 20 min intervals at progressively higher speeds until exhausted or until all flume speeds had been monitored. The number of times the fish surfaced, presumably to breathe, was recorded and the events were averaged over that 20 min period.

To monitor oxygen consumption during swimming in well-oxygenated water, the aquatic partial pressure of oxygen in the sealed flume was increased by approximately 666.5–1333 Pa (5–10 mmHg) above air-saturation by diverting some of the water from the flume into a bucket, where an air-stone was used to send bubbles of oxygen through the water. This super-saturated water was returned to the flume. Elevation of oxygen levels at the beginning of the experiment ensured that the water would be close to air-saturated during the exercise period. Attempts were made to minimize disturbance of the fish during this procedure. Once the oxygen level had been increased, oxygen consumption from air and water was measured for at least 1 h while the fish was resting. At the end of this time, the trolling motor was turned on and the *A. calva* swam for at least 1 h at a flume speed of 0.5 m s⁻¹. The *L. oculatus* swam for 1 h at a slower speed, approximately 0.3 m s⁻¹. Although data were collected for only 1 h, the fish were able to maintain these speeds for more than twice this period. After 1 h of swimming, the trolling motor was turned off and the fish was monitored for a recovery period of at least 1 h. The fish was then removed from the flume, and the experiment was repeated to control for oxygen consumption from the water by bacteria and for diffusion across the air–water interface.

Results

Preliminary experiments revealed a relationship between the frequency of breathing of *A. calva* and swimming speed. At 15 °C in well-oxygenated water, none of the six fish examined took a breath in over 1 h of observation while at rest. During exercise, the period between surfacing events grew shorter as the fish swam faster (Fig. 3). The fish surfaced more frequently at the end of the 20 min period than at the beginning. By the end of the 20 min period at the highest speeds, some fish surfaced as frequently as twice a minute. Furthermore, it was noted that fish that air-breathed frequently could sustain activity for a longer period than those that did not. For example, surfacing events were reduced at higher speeds in fish number 3. Concomitant with this reduction was a poor swimming performance, with the fish losing balance and rolling on its side, indicating it was exhausted. In contrast, after

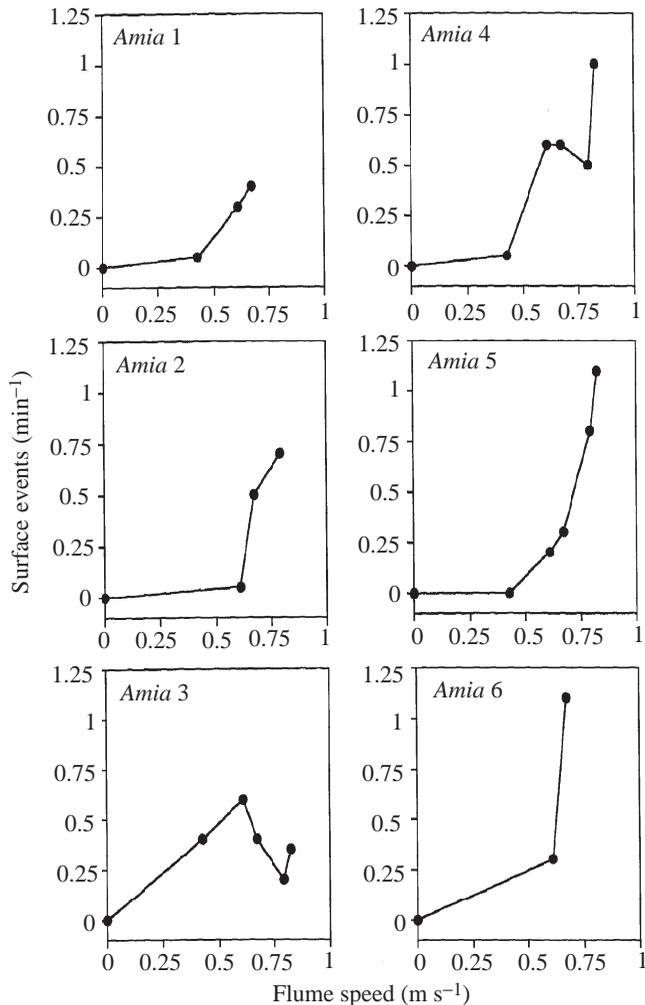


Fig. 3. The frequency of surfacing events, presumed to indicate air-breaths, as a function of flume speed for *Amia calva* swimming in an open flume at 15 °C. The fish were observed at rest for over 1 h. During this time, none of the six fish surfaced. The flume was then started at the lowest speed, and the number of times the fish surfaced during a 20 min interval was recorded and averaged over that interval. The speed was increased for another 20 min period until the fish showed signs of exhaustion or all flume speeds had been reached. In general, the faster the fish swam, the more frequently they surfaced.

swimming for 20 min at the highest speed, fish number 5 showed no signs of fatigue.

Even though they were at warmer temperatures (19–23 °C), the *A. calva* in the sealed flume rarely air-breathed at rest, consuming oxygen at an average rate of $0.81 \pm 0.314 \text{ ml O}_2 \text{ min}^{-1} \text{ kg}^{-1}$ (STD) from water and $0.15 \pm 0.331 \text{ ml O}_2 \text{ min}^{-1} \text{ kg}^{-1}$ (mean \pm S.D., $N=5$) from air. During activity, the fish often did not take a breath for the first 10–15 min of the exercise period, but then air-breathed regularly, consuming oxygen at a significantly faster rate from air ($2.65 \pm 0.708 \text{ ml O}_2 \text{ min}^{-1} \text{ kg}^{-1}$) than at rest (mean \pm S.D., $N=5$; $P \leq 0.001$; paired *t*-test). Fig. 4 shows a sample of data obtained from a single *A. calva* during an exercise trial.

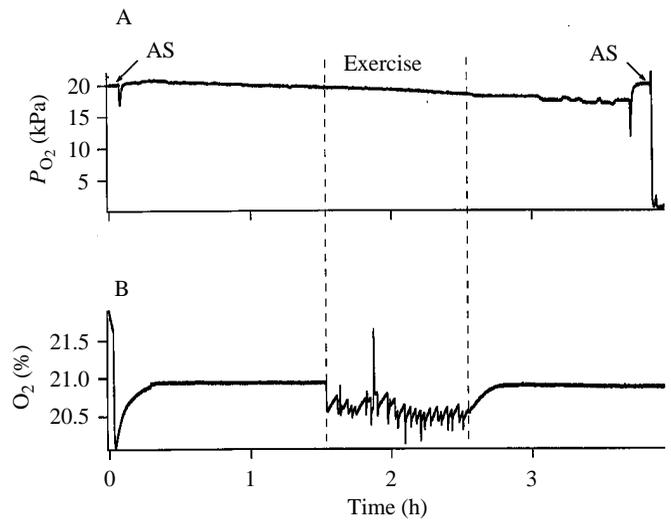


Fig. 4. Sample data from an *Amia calva* swimming in a sealed flume at 19–23 °C. (A) The partial pressure of oxygen (P_{O_2}) of the water in the flume. The regions marked AS indicate the electrode reading when air-saturated water was pumped through the electrode chamber. This water, as well as humidified nitrogen gas, was circulated before, after and sometimes during the experiments to monitor drift in the electrode. (B) The percentage oxygen in the air flowing through the air-box. The initial deflection in the trace was due to the flow control unit being turned on. The oxygen electrode was calibrated to read 20.95% with room air, and the air-hole was then sealed. The deflections of the trace during exercise correspond to air-breaths. As the fish exhaled nitrogen into the air-box, the oxygen was diluted. *A. calva* often spit out water while taking a breath (Randall *et al.* 1981). The large spike approximately one-third of the way through the exercise period is due to a particularly good shot that caused some of this water momentarily to interrupt the flow of air through the tubing.

Fig. 4A shows the aquatic partial pressure of oxygen in the flume. Fig. 4B shows the percentage of oxygen in the air-box. During the hour or so of rest that was monitored, the fish did not air-breathe. The downward deflections in Fig. 4B during the exercise period represent breaths. As nitrogen was exhaled into the air-hole, the ratio of oxygen reaching the meter decreased. None of the fish was found to air-breathe during recovery. A similar pattern of air-breathing was observed in *L. oculatus*. Resting fish consumed oxygen from the water at a mean rate of $0.62 \pm 0.067 \text{ ml O}_2 \text{ min}^{-1} \text{ kg}^{-1}$ and from air at $0.01 \pm 0.012 \text{ ml O}_2 \text{ min}^{-1} \text{ kg}^{-1}$ (mean \pm S.D., $N=4$). During activity, the rate of oxygen consumption from air ($0.87 \pm 0.720 \text{ ml O}_2 \text{ min}^{-1} \text{ kg}^{-1}$) was significantly greater than at rest ($P=0.029$; Mann–Whitney rank sum test). Data for rates of oxygen consumption from water and from air for *A. calva* and *L. oculatus* are reported in Table 1.

There was no correlation between the partial pressure of oxygen in the water and aerial oxygen consumption either during rest or exercise. For example, the *A. calva* that consumed the least amount of oxygen from air during exercise was swimming in water with a lower partial pressure of oxygen than the *A. calva* that consumed the greatest amount of oxygen

Table 1. Rates of oxygen consumption from gills and lungs at 19–23 °C

	Rest				Exercise			
	Air	Water	Total	Air %	Air	Water	Total	Air %
<i>Lepisosteus oculatus</i> 1	0.02	0.59	0.61	3	0.31	0.93	1.24	25
<i>Lepisosteus oculatus</i> 2	0.02	0.57	0.59	4	0.52	0.48	1.00	52
<i>Lepisosteus oculatus</i> 3	0	0.72	0.72	0	0.74	0.62	1.36	54
<i>Lepisosteus oculatus</i> 4	0	0.61	0.61	0	1.92	0.45	2.37	81
Mean ± S.D.				1.7±2.1				53.0±22.9
<i>Amia calva</i> 1	0	1.10	1.10	0	2.47	1.20	3.67	67
<i>Amia calva</i> 2	0	0.52	0.52	0	2.78	1.34	4.12	68
<i>Amia calva</i> 3	0	0.52	0.52	0	1.85	0.88	2.73	68
<i>Amia calva</i> 4	0	1.18	1.18	0	2.40	2.07	4.47	53
<i>Amia calva</i> 5	0.74	0.74	1.44	50	3.77	1.18	4.95	76
Mean ± S.D.				10.0±22.4				66.4±8.3

Rates of oxygen consumption are given in ml min⁻¹ kg⁻¹.

from air. The partial pressure of oxygen in the water at the beginning of the exercise period varied between 19.3 kPa (145 mmHg) and 20.7 kPa (155 mmHg), depending on the partial pressure in the water when the flume was sealed, the length of rest monitored and the rate of oxygen consumption during resting. The aquatic partial pressure of oxygen during the experiments was always greater than 17.3 kPa (130 mmHg).

Discussion

Air-breathing during exercise in oxygen-rich water is important to both *A. calva* and *L. oculatus*, while playing a smaller role during rest. Consistent with previous observations of *A. calva* at low temperatures (Johansen *et al.* 1970), these fish rarely air-breathed at rest. However, when swimming vigorously, some fish surfaced to breathe as often as twice a minute. The more intense the exercise, the more frequently the fish rose to the surface. Although increasing temperature increases the rate of air-breathing in *A. calva* (Johansen *et al.* 1970), in the present study at warm temperatures (19–23 °C), resting fish still rarely air-breathed. For only one of the five *A. calva* studied, lung ventilation provided 50% of the oxygen consumed. However, this fish had a resting rate of oxygen consumption that was considerably higher than those of the other fish studied. This may indicate that the animal was not truly resting, but was swimming in the flume. The rates of resting pulmonary oxygen consumption found for *A. calva* at 19–23 °C are in agreement with the data Randall *et al.* (1981) but are slightly lower than values reported by Johansen *et al.* (1970). *L. oculatus* also increase air-breathing with temperature (Rahn *et al.* 1971; Smatresk and Cameron, 1982a,b). However, during rest in the present study at relatively warm temperatures (19–23 °C), they extracted a minor percentage of their oxygen from air. The resting rates of pulmonary oxygen consumption reported here for *L. oculatus* are lower than those reported by Smatresk and Cameron (1982a). Although attempts were made to accustom the fish to

the flume, it is an unnatural environment that may have provoked a diving reflex. However, if a diving reflex were a factor in inhibiting air-breathing during rest, it would presumably have continued to inhibit air-breathing during activity. However, activity increased the rate of pulmonary oxygen consumption for both species; during slow swimming, the fish consumed a majority of their oxygen from the lung, rather than from the gill. Thus, the increased rate of air-breathing seen during exercise for all fish examined probably indicates an increased drive to breathe.

In a natural setting, lung ventilation in *A. calva* and *L. oculatus* may be more important in support of activity than for survival in hypoxic habitats. Unlike activity, hypoxia is not a strong stimulus to air-breathe in either genus. In numerous observations of *L. oculatus*, Rahn *et al.* (1971) found no correlation between the partial pressure of oxygen in the water and the breathing interval. Furthermore, Johansen *et al.* (1970) noted that the partial pressure of oxygen in cold (10 °C) water dropped to as low as 5.3 kPa (40 mmHg) before some *A. calva* were motivated to air-breathe. *A. calva* respond to hypoxia by increasing rates of gill ventilation until the hypoxia becomes extreme (Johansen *et al.* 1970). Of course, these fish obtain oxygen from the air when it is scarce in the water, but it is unclear how important this ability is in their natural environment. In contrast, field observations link air-breathing to times of activity, such as spawning and feeding, in both *A. calva* and *L. oculatus* (Becker, 1983; Reighard, 1903; Saksena, 1963).

The findings that these fish air-breathe when active is in keeping with similar observations of other fish: the lungfish *Neoceratodus forsteri* (Grigg, 1965), a primitive polypterid, *Polypterus senegalus* (Magid, 1966), and a primitive elopomorph, *Megalops atlanticus* (Shlaifer and Breder, 1940). However, the reason that fish air-breathe when in oxygen-rich water is not understood. Limitations of oxygen uptake through the gills and osmoregulatory stresses associated with gill ventilation could play a role (Packard, 1974; Shipman, 1989). It is also possible that air-breathing in these species (*A. calva*,

L. oculatus, *P. senegalus* and *M. atlanticus*) is important in enhancing cardiac performance by providing the heart with oxygen (Farmer, 1997).

We thank the Florida Freshwater Fish and Game Commission and the National Biological Survey for help collecting the animals. We are indebted to S. Karlsson for advice in construction of the flumes. We thank A. P. Farrell and D. R. Jones, who made the preliminary studies possible. We are indebted to D. Carrier, C. Janis and G. Ultsch for helpful conversations and critical reading of the manuscript. This work was supported by NSF grant IBN 94-20017 to D.C.J. and NSF dissertation grant IBN-9423297.

References

- BECKER, G. C. (1983). *Fishes of Wisconsin*. pp. 239–254. Madison: University of Wisconsin.
- BRAINERD, E. L. (1994). The evolution of lung–gill bimodal breathing and the homology of vertebrate respiratory pumps. *Am. Zool.* **34**, 289–299.
- DEYST, K. A. AND LIEM, K. F. (1985). The muscular basis of aerial ventilation of the primitive lung of *Amia calva*. *Respir. Physiol.* **59**, 213–223.
- FARMER, C. (1997). Did lungs and the intracardiac shunt evolve to oxygenate the heart in vertebrates? *Paleobiology* **23**, 358–372.
- GRIGG, G. (1965). Studies on the Queensland lungfish, *Neoceratodus forsteri* (Kreffft). III. Aerial respiration in relation to habits. *Aust. J. Zool.* **13**, 413–421.
- GUNTHER, A. (1871). Description of *Ceratodus*, a genus of Ganoid fishes, recently discovered in rivers of Queensland, Australia. *Phil. Trans. R. Soc. Lond.* **161**, 511–571.
- JOHANSEN, K., HANSON, D. AND LENFANT, C. (1970). Respiration in a primitive air breather, *Amia calva*. *Respir. Physiol.* **9**, 162–172.
- KRAMER, D. L., LINDSEY, C. C., MOODIE, G. E. E. AND STEVENS, E. D. (1978). The fishes and the aquatic environment of the central Amazon basin, with particular reference to respiratory patterns. *Can. J. Zool.* **56**, 717–729.
- LAUDER, G. V. AND LIEM, K. F. (1983). The evolution and interrelationships of the actinopterygian fishes. *Bull. Mus. Comp. Zool.* **150**, 95–197.
- LIEM, K. F. (1988). Form and function of lungs: The evolution of air-breathing mechanisms. *Am. Zool.* **28**, 739–759.
- MAGID, A. M. A. (1966). Breathing and function in the spiracles in *Polypterus senegalus*. *Anim. Behav.* **14**, 530–533.
- PACKARD, G. C. (1974). The evolution of air breathing in Paleozoic gnathostome fishes. *Evolution* **28**, 320–325.
- RAHN, H., RAHN, K. B., HOWELL, B. J., GANS, C. AND TENNEY, S. M. (1971). Air-breathing of the garfish (*Lepisosteus osseus*). *Respir. Physiol.* **11**, 285–307.
- RANDALL, D. J., CAMERON, J. N., DAXBOECK, C. AND SMATRESK, N. (1981). Aspects of bimodal gas exchange in the bowfin, *Amia calva* L. (Actinopterygii: Amiiformes). *Respir. Physiol.* **43**, 339–348.
- REIGHARD, J. (1903). The natural history of *Amia calva* Linnaeus. In *Mark Anniversary Volume*, pp. 59–109. New York: Henry Holt and Co.
- SAKSENA, V. P. (1963). Effects of temperature, light, feeding and activity on the rate of aerial breathing in gar (*Lepisosteus*). *Diss. Abstr.* **24**, 2628.
- SHIPMAN, B. (1989). Patterns of ventilation and acid–base recovery following exhausting activity in the air-breathing fish *Lepisosteus oculatus*. MSc thesis, University of Texas, Arlington, USA.
- SHLAIFER, A. AND BREDER, C. M. (1940). Social and respiratory behavior of small Tarpon. *Zoologica* **25**, 493–512.
- SMATRESK, N. J. AND CAMERON, J. N. (1982a). Respiration and acid–base physiology of the spotted gar, a bimodal breather. I. Normal values and the response to severe hypoxia. *J. exp. Biol.* **96**, 263–280.
- SMATRESK, N. J. AND CAMERON, J. N. (1982b). Respiration and acid–base physiology of the spotted gar, a bimodal breather. II. Responses to temperature change and hypercapnia. *J. exp. Biol.* **96**, 281–293.