The Journal of Experimental Biology 203, 1671–1678 (2000)
Printed in Great Britain © The Company of Biologists Limited 2000
JEB2067

VENTILATION AND GAS EXCHANGE DURING TREADMILL LOCOMOTION IN THE AMERICAN ALLIGATOR (*ALLIGATOR MISSISSIPPIENSIS*)

COLLEEN G. FARMER[1,*] AND DAVID R. CARRIER[2]

[1]*Department of Ecology and Evolutionary Biology, 321 Steinhaus Hall, University of California, Irvine, CA 92697, USA* and [2]*Department of Biology, University of Utah, 257 South 1400 East, Salt Lake City, UT 84112-0840, USA*

*e-mail: cfarmer@uci.edu

Accepted 22 March; published on WWW 10 May 2000

Summary

A number of anatomical characters of crocodilians appear to be inconsistent with their lifestyle as sit-and-wait predators. To address this paradoxical association of characters further, we measured lung ventilation and respiratory gas exchange during walking in American alligators (*Alligator mississippiensis*). During exercise, ventilation consisted of low-frequency, large-volume breaths. The alligators hyperventilated severely during walking with respect to their metabolic demands. Air convection requirements were among the highest and estimates of lung P_{CO_2} were among the lowest known in air-breathing vertebrates. Air convection requirements dropped immediately with cessation of exercise. These observations indicate that the ventilation of alligators is not limited by their locomotor movements. We suggest that the highly specialized ventilatory system of modern crocodilians represents a legacy from cursorial ancestors rather than an adaptation to a lifestyle as amphibious sit-and-wait predators.

Key words: alligator, *Alligator mississippiensis*, ventilation, oxygen consumption, carbon dioxide excretion, locomotion, hyperventilation.

Introduction

The anatomy of crocodilians is paradoxical. Aspects of their locomotor and cardiac anatomy seem to be more consistent with the vigorous activity levels of many birds and mammals than with their lifestyle as sit-and-wait predators. The nearly parasagittal limb posture they exhibit when walking, the bounding gait observed in the young of some species, the diaphragmatic muscle (which may help them to breathe effectively during locomotion) and a cardiac ventricle that enables elevated pressure in the systemic circulation without elevated pulmonary pressures are all features that are usually associated with high levels of sustained activity (Carrier, 1987a; Farmer, 1999; Walker, 1970). Nevertheless, crocodilians have a poor capacity for vigorous, sustained, terrestrial locomotion.

Maintenance of vigorous activity is made possible through high levels of aerobic metabolism which are dependent on high rates of respiratory gas exchange. Because the respiratory, cardiac and peripheral circulatory systems function in an integrated fashion to transport oxygen and carbon dioxide (Janicki et al., 1996), the question arises as to whether the ventilatory system of crocodilians is suited to their sit-and-wait lifestyle or whether it matches the perplexing design of their locomotor and cardiovascular systems. Furthermore, a ventilatory bottleneck for gas exchange was predicted to occur during locomotion in animals that have a sprawling posture (Carrier, 1987a,b). Later studies confirmed that low rates of costal ventilation during locomotion can limit gas exchange for lepidosaurs with a sprawling posture (Owerkowicz et al., 1999; Wang et al., 1997). Furthermore, on the basis of anatomical and locomotor features, Carrier (1987a) predicted that crocodilians would not experience this mechanical constraint. Consequently, we examined whether ventilation in a crocodilian is constrained during terrestrial locomotion. This study investigates aspects of the respiratory physiology of *Alligator mississippiensis* in an attempt to uncover similarities and differences between crocodilians and other amniotes.

Materials and methods

Animals

American alligators (*Alligator mississippiensis*) were kept in aquaria with basking platforms and both heat and full-spectrum light sources. They experienced a photoperiod of 14 h:10 h light:dark. They were fed a diet of goldfish, smelts, mice, rats and eggs. The mass of the animals ranged from 1.18 to 1.66 kg, and the mean mass of the five animals used was 1.34 kg.

Training

Animals were trained to walk on a treadmill over the course of several months. Although the animals initially had difficulty sustaining locomotion, by exposing them to short training sessions (several minutes of walking with 10 min intervals for rest) 2–3 times a week, their endurance improved until they could sustain repeated bouts of 4 min of continuous walking interspersed with 25 min of rest.

Ventilation and gas exchange

To measure ventilation and gas exchange, a mask was constructed out of the tip of a 30 ml plastic syringe. Two ports were drilled into the syringe, and flexible Hytrel tubing (Hans Rudolph, Inc., Kansas City, MO, USA) with an inner diameter of 9 mm was glued to the ports. Flow control units (Ametek, Pittsburgh, PA, USA) were used to pull fresh air ($F_{\mathrm{I}O_2}$=0.2093 and $F_{\mathrm{I}CO_2}$=0.0003, where $F_{\mathrm{I}O_2}$ and $F_{\mathrm{I}CO_2}$ are the fractional gas concentrations in the inspired air) through the mask at an approximate rate of 5.4 l min^{-1}. This was the biased flow determined to eliminate rebreathing by the alligators during their most vigorous exercise. A pneumotachometer (8311 Hans Rudolph, Inc., Kansas City, MO, USA) was placed in the line upstream from the mask. A portion of the gas flowing through the mask was diverted and pulled through Drierite (anhydrous calcium sulfate) and then through oxygen (Beckman OM-11, Fullerton, CA, USA) and carbon dioxide (Ametek CD-3A, Pittsburgh, PA, USA) analyzers. The mask was sealed with epoxy adhesive over the nares. Preliminary experiments revealed that alligators breathe through the mouth as well as through the nares, especially when vocalizing during exercise. Hence, the mouth of the animal was sealed with duct tape to ensure that all respiratory gases were collected. The ventilation and gas-exchange system was calibrated by injecting known volumes of gas into the mask before it was attached to the animal.

A separate arrangement was used to measure standard metabolic rate. An animal was enclosed in a respirometer constructed from airtight plastic. Air was pulled through the container, then through a flowmeter (Gilmont Instruments, Barrington, IL, USA), into a tube containing Drierite and finally into the oxygen and carbon dioxide analyzers. The animal was left undisturbed overnight before data were collected the following morning.

Stride

A mercury strain gauge, wired to one arm of a Wheatstone bridge, was mounted to the back of each animal using Velcro. Bending of the trunk during walking stretched the gauge, altered the resistance of the mercury column and thus provided an indication of the locomotor cycle.

Data collection and analysis

Analog signals from the gas analyzers, the strain gauge and the differential pressure transducer of the pneumotach were converted to digital format using an A/D converter (Biopack System, Goleta, CA, USA) and stored on a Macintosh computer. Signals were sampled at a rate of 50 Hz and analyzed with AcqKnowledge software (Biopack System, Goleta, CA, USA).

Ventilation is reported at body temperature, ambient pressure and saturated with water vapor (BTPS). Gas exchange and estimates of blood gases are expressed at standard temperature and pressure, and for dry gas (STPD) (West, 1979). Pre-exercise values for minute ventilation ($\dot{V}_\mathrm{E}$), tidal volume, respiratory frequency and rates of oxygen consumption ($\dot{V}_{O_2}$) and carbon dioxide excretion ($\dot{V}_{CO_2}$) were obtained several hours after the animal had been instrumented. Exercise values for oxygen consumption, carbon dioxide excretion, minute ventilation, respiratory frequency and tidal volume were collected during the last minute of 4 min bouts of exercise. Similarly, these data were collected during the first minute of recovery after exercise, starting with the first breath after the exercise period. These values were used to compute respiratory gas exchange ratios (RER; $\dot{V}_{CO_2}/\dot{V}_{O_2}$), air convection requirements ($\dot{V}_\mathrm{E}/\dot{V}_{O_2}$ and $\dot{V}_\mathrm{E}/\dot{V}_{CO_2}$) and extraction ($\dot{V}_{O_2}/\dot{V}_\mathrm{E}\times F_{\mathrm{I}O_2}$) and to estimate faveolar P_{CO_2} (STPD) using the alveolar gas equation assuming a dead space volume of 4.2 ml kg^{-1} (Hicks and White, 1992). Furthermore, comparison of the air convection requirements before, during and after exercise allowed us to assess whether the animals were hyper- or hypoventilating. We use the terms to mean a change in lung ventilation that results in a reduction or increase, respectively, in the partial pressure of carbon dioxide within the lung compared with pre-exercise conditions.

The duration of inspiration and expiration for each breath was measured during pre-exercise, during exercise at a speed of 0.31 m s^{-1} and for the first minute of recovery from exercise at 0.31 m s^{-1}. The volume of each breath was then divided by the time it took to either inspire or expire to give mean inspiratory and expiratory flows.

The Fick principle was used to calculate rates of oxygen consumption and carbon dioxide excretion of four animals in the respirometer. Oxygen consumption or carbon dioxide excretion is equal to the difference between the fraction of air flowing into the box that is oxygen ($F_{\mathrm{I}O_2}$) or carbon dioxide ($F_{\mathrm{I}CO_2}$) and the fraction of air flowing out of the box that is oxygen or carbon dioxide multiplied by the flow of air.

Experimental protocol

All animals had fasted for at least 4 days before the experiments. They were brought from the animal care facility to the experimental chamber the night before data were collected. The experimental chamber was kept at 30 °C. After instrumenting, an animal it was given several hours on the treadmill to calm down from the handling before pre-exercise data were collected. The treadmill was then started, and the animal exercised for at least 3 min but not more than 4 min. This period provided 1 min of data after the walking alligators had reached steady state, which occurred within 1.5–2 min after the start of exercise. After 4 min, the treadmill was stopped and a period of recovery was monitored. After no less than 25 min of rest, data were collected from a second exercise trial at another speed. This was the period required for ventilation (both tidal volume and respiratory frequency) to return to pre-exercise levels (Farmer and Carrier, 2000b). The

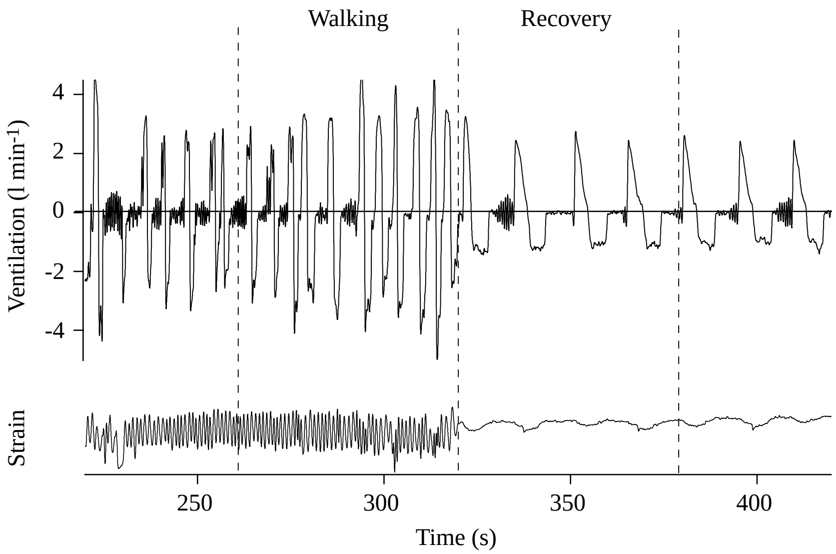

Fig. 1. Sample data of ventilation during walking in an American alligator illustrating the exercise/recovery transition. The top trace shows the signal from the pneumotachometer; exhalation occurs when the values are positive. The high-frequency ventilatory signals that sometimes occur during apnea are due to movement of air into and out of the buccal area. This air-flow did not contribute to lung ventilation and was not included in the analysis of ventilation. The periods between the vertical dashed lines represent the last minute of exercise and the first minute of recovery. A dramatic decrease in ventilatory frequency is apparent upon cessation of exercise. The bottom trace is the signal from the strain gauge. Each wave cycle indicates one locomotor cycle. The constant frequency and amplitude of the oscillations in this trace are indicative of steady locomotion. Large breaths occur during walking. The small-amplitude oscillations during recovery are due to motion of the thorax and abdomen during ventilation, not to locomotion.

protocol consisted of a graded increase in exercise. The speeds were not randomized. All data were collected on the same day.

Standard metabolic rates were determined on a separate day, at least 2 days after a training or experimental exercise trial. An animal was sealed in a respirometer, and air was pulled through the respirometer at a rate of 250 ml min^{-1}. The animal was left undisturbed overnight before data were collected the following day. The air passed through Drierite before being sampled by the oxygen and carbon dioxide meters. Monitoring continued for at least 2 h. Periodic checks of the analyzers were made by switching a valve to pump fresh air through the analyzer for short periods to account for drift in the meters.

Results

The pattern of ventilation varied abruptly with the transition from pre-exercise to exercise and with the transition from exercise to recovery from exercise. Fig. 1 illustrates the transition that occurred upon cessation of exercise. Minute ventilation increased from 19.3±3.1 ml BTPS min^{-1} kg^{-1} before exercise to 1199.7±332.9 ml min^{-1} kg^{-1} (means ± s.e.m., N=5) during locomotion at 0.44 m s^{-1} (Fig. 2). This increase was due both to an increase in tidal volume, from 21.7±2.9 ml kg^{-1} before exercise to 84.7±12.3 ml kg^{-1} during exercise, and to an increase in respiratory frequency, from 0.92±0.12 breaths min^{-1} before exercise to 13.63±2.40 breaths min^{-1}, at a speed of 0.44 m s^{-1} (Fig. 2). Minute ventilation decreased to 346±44.0 ml min^{-1} kg^{-1} upon cessation of exercise at 0.44 m s^{-1} because of an insignificant decrease in tidal volume to 82.7±7.4 ml kg^{-1} and a significant decrease in respiratory frequency to 4.17±0.29 breath min^{-1} (Fig. 2).

Ventilation during locomotion in tetrapods has often been observed to be correlated temporally with locomotor movements in one fashion or another. In this study, stride was monitored using the mercury strain gauge attached to the animal's back, and no temporal correlation was found between the phases of the ventilatory cycles and the locomotor cycles. Furthermore, no correlation was found between breaths of large volume and brief pauses in the locomotor movements. However, exercise influenced both the breath duration and the mean flow. The duration of inspiration and expiration before, during and after exercise at 0.31 m s^{-1} is reported in Table 1. Breaths before exercise were highly variable in duration and lasted 9.1±3.1 s. Mean expiratory flow was 369±98 ml min^{-1} kg^{-1}. During exercise, mean breath duration decreased to 3.3±0.2 s and expiratory flow increased to 3242±462 ml min^{-1} kg^{-1}. After exercise, the duration of the breaths increased to 6.0±0.4 s and mean expiratory flow decreased to 1433±133 ml min^{-1} kg^{-1}.

The rate of oxygen consumption ($\dot{V}_{O_2}$) increased with increasing locomotor speed (Fig. 2). During the first minute of recovery, the rate of oxygen consumption was less than or equal to that during the last minute of exercise (Fig. 2). The rate of carbon dioxide excretion ($\dot{V}_{CO_2}$) also increased with increasing locomotor speed (Fig. 2). The respiratory exchange ratio increased from 0.7±0.04 before exercise to 1.4±0.06

Table 1. *Inspiratory and expiratory durations and mean air flow*

	Pre-exercise	Exercise at 0.31 m s^{-1}	Recovery
Inspiratory flow (ml BTPS min^{-1} kg^{-1})	370±83	2970±517	1374±170
Expiratory flow (ml BTPS min^{-1} kg^{-1})	369±98	3242±462	1433±133
T_{tot} (s)	9.1±3.1	3.3±0.2	6.0±0.4
T_I (s)	4.2±1.1	1.7±0.1	3.1±0.3
T_E (s)	4.9±2.0	1.6±0.1	2.9±0.1

Values are means ± s.e.m., N=5.

T_I, inspiratory duration; T_E, expiratory duration; T_{tot}, total breath duration.

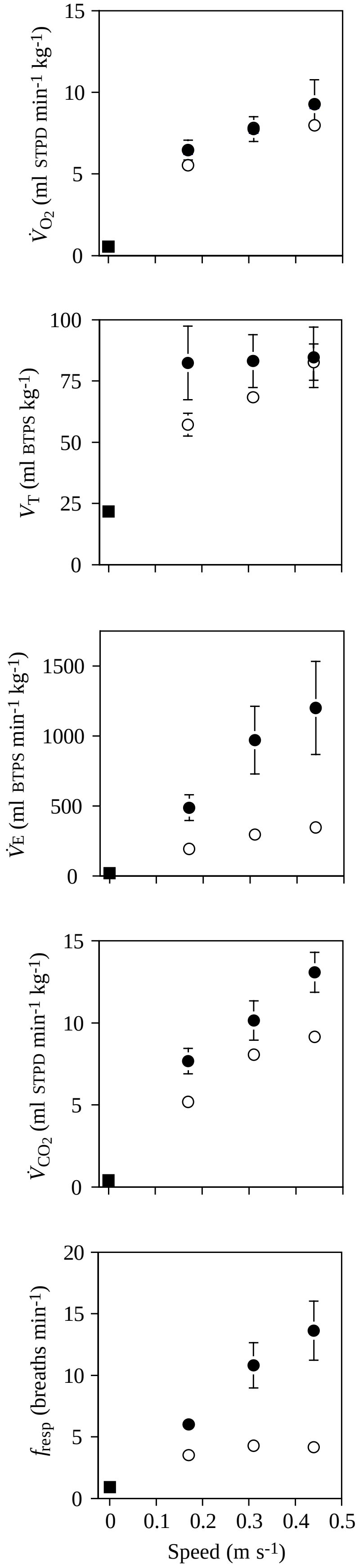

Fig. 2. The means and standard error about the mean for rates of oxygen consumption ($\dot{V}_{\mathrm{O}_2}$) and carbon dioxide excretion ($\dot{V}_{\mathrm{CO}_2}$), minute ventilation ($\dot{V}_{\mathrm{E}}$), tidal volume (V_{T}) and respiratory frequency (f_{resp}) for five alligators before exercise, during the last minute of exercise at various treadmill speeds and during the first minute of recovery. If error bars are not seen, it is because they are contained within the symbols. Pre-exercise, filled squares; exercise, filled circles; recovery from exercise, open circles.

during exercise (Fig. 3). The ratio decreased during the first minute of recovery (Fig. 3).

Air convection requirements for oxygen ($\dot{V}_{\mathrm{E}}/\dot{V}_{\mathrm{O}_2}$) increased from pre-exercise values of 34.9±3.3 to 127.6±29.5 during exercise at 0.44 m s^{-1} (Fig. 3). This ratio decreased immediately during the first minute of recovery to 43.5±5.3. Air convection requirements for carbon dioxide also increased from pre-exercise values of 48.8±2.5 to 89.2±18.9 during exercise at 0.44 m s^{-1}. This ratio also decreased immediately during recovery to 34.0±4.1 (Fig. 3).

Estimates of falveolar P_{CO_2} declined from pre-exercise values of 2.1±0.1 STPD kPa to 1.1±0.3 kPa during exercise at 0.44 m s^{-1}. Recovery values immediately increased to pre-exercise levels (Fig. 3). Estimates of extraction declined from a pre-exercise value of 0.19±0.018 to 0.06±0.017 during exercise at 0.44 m s^{-1} (Fig. 3). Extraction immediately increased during recovery to 0.15±0.022.

The standard rate of oxygen consumption for the four animals studied was 0.30±0.02 ml STPD min^{-1} kg^{-1} (mean ± S.E.M.). Carbon dioxide excretion was 0.23±0.017 ml STPD min^{-1} kg^{-1} (mean ± S.E.M.) and respiratory quotient (RQ) was 0.77±0.018 (mean ± S.E.M.).

Discussion

Critique of the methods

We studied immature and captive-raised animals while exercising on a treadmill. Locomotion on a treadmill is an unnatural situation. Consequently, it is possible that the response to exercise with this protocol would be different in adult animals and may not reflect the response of wild animals living in natural environments.

Another caveat concerns the effects of training. If our training regimen was not rigorous enough, we may be reporting values that are considerably lower than would be found in fit wild animals. However, if our training regimen was too vigorous, we might have conditioned the animals to a level of performance that is greater than would be found in the wild. We may have changed a number of parameters, e.g. strengthened locomotor muscles, increased the capacitance of the muscles for oxygen uptake, changed circulatory or acid–base variables and changed the ventilatory response to exercise.

Because no data have been collected on oxygen consumption, carbon dioxide excretion and ventilation from wild animals exercising under natural conditions, it is impossible to assess whether our training regimen produced an artificial and unnatural response to exercise. However, we can compare our data with those collected by other researchers in laboratory

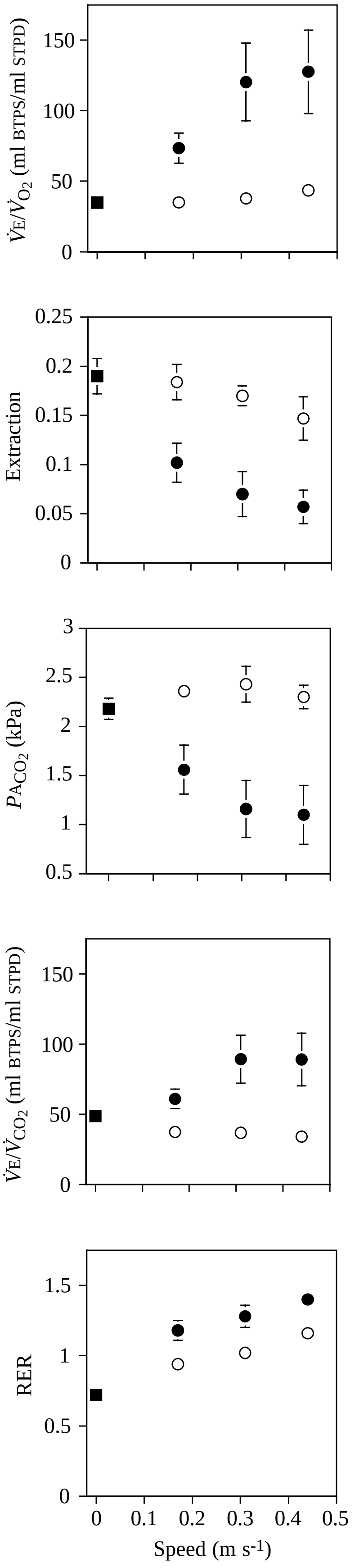

Fig. 3. The means and standard error for air convection requirements ($\dot{V}_E/\dot{V}_{O_2}$ and $\dot{V}_E/\dot{V}_{CO_2}$), extraction, respiratory exchange ratio (RER) and estimates of faveolar P_{CO_2} ($P_{A_{CO_2}}$) calculated using the alveolar gas equation for five alligators before exercise, for the last minute of exercise at various treadmill speeds and during the first minute of recovery. If error bars are not seen, it is because they are contained within the symbols. Pre-exercise, filled squares; exercise, filled circles; recovery from exercise, open circles.

environments. Maximal rates of oxygen consumption ($\dot{V}_{O_2max}$) during treadmill exercise at 30 °C in American alligators weighing 249–352 g have been reported to be approximately 3.7 ml min^{-1} kg^{-1} (values are not converted to STPD) (Emshwiller and Gleeson, 1997). Although we do not claim to have measured $\dot{V}_{O_2max}$, our values recorded at 0.44 m s^{-1} were approximately three times higher (from significantly larger animals). Furthermore, maximal struggling activity in *Crocodylus porosus* elicited an oxygen consumption rate of only 2.8 ml min^{-1} kg^{-1} (Wright, 1986). This suggests that, compared with other studies, we were not working with unhealthy animals that were somehow impaired by captivity or training.

We wish to emphasize that other modes of exercise (e.g. swimming) may produce different results. It is well known that the maximum rates of oxygen consumption elicited from humans when running are greater than during bicycle exercise. Similarly, aquatic locomotion in alligators may yield different values for oxygen consumption from terrestrial locomotion. Indeed, there are some data showing that this is probably the case. Lewis and Gatten (1985) report that alligators exercising in water had lower rates of oxygen consumption than during terrestrial locomotion.

Another important caveat concerns the limitations of our repeat protocol. It is well established that the effects of exercise on physiological variables can extend well beyond the exercise period. For example, depending on the intensity of exercise, oxygen consumption measurements were elevated in juvenile alligators from approximately 2.5 to over 5 h after exercise ceased (Hernandez and Coulson, 1980). Similarly, in humans, it can take as long as 48–56 h for oxygen consumption measurements to return to pre-exercise values (Bullough et al., 1995). Hence, previous exercise bouts may have influenced physiological variables in subsequent exercise periods. For example, the rate of oxygen consumption at 0.44 m s^{-1} might be elevated because the animal was walking at a faster pace or because of the previous exercise bout, or because of both the previous exercise bout and the more strenuous pace. It is impossible to distinguish from our data the relative contributions of these factors to the value we report. Although this protocol has limitations, it has been used to study the response to exercise of other species (Owerkowicz et al., 1999; Wang et al., 1997). Hence, we feel that comparisons of these data with data on other species collected under similar conditions, repeated bouts of exercise, have produced internally consistent results.

Ventilatory pattern

Lung ventilation of resting American alligators is typical of other ectothermic tetrapods, consisting of intermittent breaths of moderate tidal volume. The resting tidal volumes we recorded, 21.7±2.9 ml kg^{-1}, are comparable with the value of 21.8±6.9 ml kg^{-1} measured by Hicks and White (1992). During treadmill exercise, however, the ventilatory pattern of alligators is strikingly different from that reported for lizards. Tidal volumes increased with locomotor speed, reaching 85 ml kg^{-1} during walking at 0.44 m s^{-1}. These breaths were approximately four times greater in volume than they were before exercise. As far as we know, these tidal volumes are the largest reported for any animal during exercise.

These results differ markedly from the ventilatory pattern of lizards. During treadmill exercise in lizards, costal tidal volumes are generally the same or smaller than those at rest (Carrier, 1987b; Wang et al., 1997). Hence, increases in minute ventilation during exercise are necessarily accomplished by increases in breathing frequency. For example, during moderate-speed locomotion in *Iguana iguana*, a doubling of minute ventilation above resting values is due to a greater than threefold increase in ventilation frequency and a 45 % reduction in tidal volume. During recovery from exercise in these lizards, tidal volume increases above exercise values by two- to fourfold. Hence, costal ventilation of lizards differs from that of alligators in that it consists of breaths of very small tidal volume during exercise and an increase in tidal volume during recovery from exercise.

Some lizards supplement costal ventilation during locomotion by pumping air into the lung by compression of the gular or buccal area (Owerkowicz et al., 1999). Although alligators frequently move air into and out of the buccal cavity, this air was found not to contribute to gas exchange. This observation is consistent with the results of previous research (Gans and Clark, 1976; Huggins et al., 1968; Naifeh et al., 1970a,b, 1971). In this regard, ventilation during exercise in alligators was also found to differ from that of some lepidosaurs.

The ventilatory pattern of alligators is also distinct from the typical mammalian pattern. In mammals, increases in ventilation during exercise are largely the result of increases in breathing frequency (influenced by the use of ventilation for evaporative cooling and by locomotor ventilatory coupling, see below). Humans are somewhat of an exception to this generalization. Normal young humans generally show an increase in tidal volume of three- to fivefold during exercise and an increase in breathing frequency of one- to threefold; recovery from exercise often consists of an increase in breathing frequency compared with exercise but a decrease in tidal volume, resulting in very rapid, shallow breathing (for a review, see Dempsey et al., 1996).

Changes in minute ventilation during flight in birds are variable. Increases in tidal volume with exercise range from approximately one- to fourfold, while increases in breathing frequency range from approximately two- to 20-fold (for reviews, see Butler, 1991; Saunders and Fedde, 1994). Whether increases in minute ventilation are accomplished primarily by changes in frequency or tidal volume or both is related to the relationship between respiratory and locomotor cycles (Boggs, 1997).

In summary, the increase in tidal volume observed in alligators during exercise is similar to the pattern found in some birds and in humans, while it contrasts with the small, costal tidal volumes of lizards and many quadrupedal mammals. Several reasons for these different ventilatory patterns can be suggested. First, ventilation often plays a role in evaporative cooling in birds and many mammals, and panting is composed of rapid shallow breaths. Second, in many mammals and birds, ventilation is often integrated with the locomotor cycle so that when stride frequency increases ventilatory frequency must also increase (Attenburrow, 1982; Boggs, 1997; Bramble and Carrier, 1983). This obligate increase in respiratory frequency is often associated with a concomitant decrease in tidal volume. Finally, because lizards have a mechanical constraint on simultaneous costal ventilation and locomotion, costal tidal volumes tend to decrease as locomotor speed increases (Carrier, 1987b).

Metabolic rate

Although minute ventilation and rates of oxygen consumption and carbon dioxide excretion increased steadily as the locomotion of the alligators became more intense, minute ventilation increased disproportionately, resulting in greatly elevated air convection requirements. Before exercise, the ratio of minute ventilation to the rate of oxygen consumption was 34.9±3.3 (mean ± S.E.M.) but this increased to 127.6±29.5 at a treadmill speed of 0.44 m s^{-1}. Air convection requirements for carbon dioxide excretion also increased. Hyperventilation during exercise is known to occur in many animals. Lizards that rely solely on costal ventilation, e.g. *Iguana iguana*, are among the few species that do not show an elevation in air convection requirements during exercise (Wang et al., 1997). Although traditional wisdom has attributed exercise hyperventilation to an exercise lactacidosis (i.e. a respiratory alkalosis has been proposed to compensate for a metabolic acidosis), recent data suggest that blood acid–base status cannot account for this phenomenon (for a review, see Forster and Pan, 1988). Hyperventilation may serve other functions, for example, to widen alveolar–arterial O_2 gradients to facilitate the saturation of arterial blood (for a review, see Forster and Pan, 1988) or to facilitate venous return (Farmer et al., 1996; Farmer and Hicks, 2000).

Because minute ventilation increased out of proportion to metabolic demand, estimates of faveolar P_{CO_2} during exercise were very low. These values fell from a mean of 2.1±0.1 STPD kPa before exercise to 1.1±0.3 kPa during exercise. If the arterial blood came to equilibrium with the gases in the lung, then arterial P_{CO_2} dropped to this low value of 1.1 kPa. Although exceptionally low compared with the values found in most air-breathing vertebrates, this value is consistent with arterial P_{CO_2} measured in *Crocodylus porosus* (Grigg and Johansen, 1987), which ranged as low as 1.4 kPa (STPD) in resting animals.

Another consequence of the large increase in air convection requirements in alligators was a significant decrease in extraction. Before exercise, extraction was 0.19±0.018. However, during exercise at 0.44 m s^{-1}, extraction decreased to 0.06±0.017. This is considerably lower than the values of

0.10–0.19 measured in exercising birds, of 0.10 measured in flying bats and of 0.12–0.22 measured in running dogs and humans (Thomas, 1981; and references therein).

The rate of oxygen consumption increased with locomotion to 9.3±0.8 ml STPD $\mathrm{min}^{-1}\,\mathrm{kg}^{-1}$, which was 17 times greater than pre-exercise values (given in Fig. 2) and 31 times greater than the standard rates. This large increase in rates of oxygen consumption over standard metabolic rates is similar and consistent with the range of values measured in other animals. For example, aerobic scopes ($\dot{V}_{\mathrm{O_2max}}/\dot{V}_{\mathrm{O_2standard}}$) in other reptiles range from 5 to 30 (Bennett, 1982), in amphibians between 6 and 26 (Taigen et al., 1982), in birds from approximately 5 to 36 (Bundle et al., 1999; Lasiewski, 1963; Tucker, 1968) and in mammals (the group that has been best studied) from approximately 3 to 60 (Morrison et al., 1959; Weibel et al., 1992). Because the values measured in our study may not reflect the full aerobic potential of American alligators (see critique of the methods), we are not suggesting that aerobic scopes are limited to 31. Yet this factorial increase is within the range measured on other tetrapods.

The absolute rates of oxygen consumption for alligators measured in this study are the largest we know of for any crocodilian during exercise. Even studies alleging to have obtained $\dot{V}_{\mathrm{O_2max}}$ from treadmill locomotion in the American alligator report values that are roughly one-third of our measurements (Emshwiller and Gleeson, 1997). Despite our relatively high values compared with other studies on crocodilians, the value of 9.3 ml STPD $\mathrm{min}^{-1}\,\mathrm{kg}^{-1}$ is rather low compared with those of other ectotherms such as the lizard *Varanus exanthematicus*, e.g. 30 ml $\mathrm{min}^{-1}\,\mathrm{kg}^{-1}$ (Owerkowicz et al., 1999). There may be several reasons for these low values.

Although 0.44 m s^{-1} was the highest speed for which we were able to maintain sustained and steady locomotion from these animals, they are capable of greater speeds. Perhaps with more training or with data collected from a track instead of a treadmill, faster locomotion and greater rates of oxygen consumption may be found. Another factor that may explain the low rates of oxygen consumption is that crocodilians are aquatic animals and their leg musculature constitutes a small fraction of their total musculature. Hence, if the tail and trunk muscles were vigorously employed, as during swimming, rates of oxygen consumption might increase significantly. However, it should be pointed out that the low rates of oxygen consumption during exercise measured in this study are very typical for animals that are sit-and-wait predators (Taigen et al., 1982; Taigen and Pough, 1983). Hence, it would be consistent with their lifestyle to find that the values we measured are near $\dot{V}_{\mathrm{O_2max}}$.

Evolutionary implications

The results of this study suggest that, unlike lizards, crocodilians do not have a mechanical constraint on simultaneous costal ventilation and locomotion. Yet, compared with other ectothermic animals, the anatomical diffusion capacity of the lung of crocodilians is very low (Perry, 1993). They have an unusually large barrier to diffusion of oxygen between the air and blood, and they have a comparatively small respiratory surface area.

One explanation for this paradoxical assemblage of characters is that extant crocodilians may have inherited a cardio-pulmonary system from ancestors that were specialized for the ability to sustain vigorous locomotion (Carrier, 1987a). Early crocodylomorphs appear to have been fully terrestrial. In the Triassic period (roughly 245 million years ago), they were slender animals about the size of a large cat and appear to have been active terrestrial predators. They had a parasagittal limb posture, long and slender limbs with a reduced number of digits, and they ran on the balls of their feet (Walker, 1970). However, by the Cretaceous period (roughly 145 million years ago), the lineage had become amphibious with anatomical features associated with an aquatic existence (e.g. a dorso-ventrally flattened skull, short limbs and expanded tail) (Pough et al., 1999). Nevertheless, most of the specialized features found in modern crocodilians evolved in the Triassic in the highly terrestrial forms (Carroll, 1988). Among birds and mammals, these characters (e.g. a parasagittal posture, thin slender limbs and reduced digits) are generally associated with a capacity for stamina.

Other observations also suggest that the evolution of locomotor stamina may have occurred early in this lineage. Early crocodylomophs had highly specialized features of the pelvis that have been found in modern forms to facilitate ventilation (Carrier and Farmer, 2000a,b; Farmer and Carrier, 2000a). Furthermore, similarities in the embryology of the avian and crocodilian ventricle suggest that a complete ventricular septum evolved early in the archosaurs (Holmes, 1975). Hence, it is possible that aspects of the highly specialized heart and ventilatory systems of modern crocodilians represent a legacy from their ancestors, rather than adaptations to their modern lifestyle as amphibious, sit-and-wait predators.

We are grateful to J. W. Hicks, K. Packard, G. Packard and to the Florida Freshwater Fish and Game Commission for supplying us with alligators. We thank J. Tlachac for his assistance with training the animals and analyzing data. We are indebted to T. Owerkowicz for comments on the manuscript and especially to J. W. Hicks for many insights, for comments on the manuscript and for the use of equipment. This study was funded by NSF IBN-9807534 to D.R.C. and NIH 1F32-HL09796-01 to C.G.F.

References

Attenburrow, D. P. (1982). Time relationship between the respiratory cycle and limb cycle in the horse. *Equine Vet. J.* **14**, 69–72.

Bennett, A. F. (1982). The energetics of reptilian activity. In *Biology of the Reptilia*, vol. 13, *Physiology D, Physiological Ecology* (ed. C. Gans and F. H. Pough), pp. 155–199. London: Academic Press.

Boggs, D. F. (1997). The effects of the wingbeat cycle on respiration in black-billed magpies (*Pica pica*). *J. Exp. Biol.* **200**, 1403–1412.

Bramble, D. M. and Carrier, D. R. (1983). Running and breathing in mammals. *Science* **219**, 251–256.

Bullough, R. C., Gillett, C. A., Harris, M. A. and Melby, C. L. (1995). Interaction of acute changes in exercise energy expenditure and energy intake on resting metabolic rate. *Am. J. Clin. Nutr.* **61**, 473–481.

Bundle, M. W., Hoppeler, H., Vock, R., Tester, J. M. and Weyand, P. G. (1999). High metabolic rates in running birds. *Nature* **387**, 31–32.

Butler, P. J. (1991). Exercise in birds. *J. Exp. Biol.* **160**, 233–262.

Carrier, D. R. (1987a). The evolution of locomotor stamina in tetrapods: circumventing a mechanical constraint. *Paleobiology* **13**, 326–341.

Carrier, D. R. (1987b). Lung ventilation during walking and running in four species of lizards. *Exp. Biol.* **47**, 33–42.

Carrier, D. R. and Farmer, C. G. (2000a). The evolution of pelvic aspiration in archosaurs. *Paleobiology* **26**, 271–293.

Carrier, D. R. and Farmer, C. G. (2000b). The integration of ventilation and locomotion in archosaurs. *Amer. Zool.*, in press.

Carroll, R. L. (1988). *Vertebrate Paleontology and Evolution*. New York: W. H. Freeman. 698pp.

Dempsey, J. A., Adams, L., Ainsworth, D. M., Fregosi, R. F., Gallagher, C. G., Guz, A., Johnson, B. D. and Powers, S. K. (1996). Airway, lung and respiratory muscle function during exercise. In *Handbook of Physiology*, section 12, *Exercise: Regulation and Integration of Multiple Systems*, vol. 12 (ed. L. B. Rowell and J. T. Shepherd), pp. 448–514. New York: Oxford University Press.

Emshwiller, M. G. and Gleeson, T. T. (1997). Temperature effects on aerobic metabolism and terrestrial locomotion in American alligators. *J. Herpetol.* **31**, 142–147.

Farmer, C. G. (1999). The evolution of the vertebrate cardio-pulmonary system. *Annu. Rev. Physiol.* **61**, 573–592.

Farmer, C. G. and Carrier, D. R. (2000a). Pelvic aspiration in the American alligator (*Alligator mississippiensis*). *J. Exp. Biol.* **203**, 1679–1687.

Farmer, C. G. and Carrier, D. R. (2000b). Respiration and gas exchange during recovery in alligators. *Respir. Physiol.* **120**, 67–73.

Farmer, C. G. and Hicks, J. W. (2000). Circulatory impairment induced by exercise in the lizard *Iguana iguana*. *J. Exp. Biol.* **203**, in press.

Farmer, C. G., Hicks, J. W. and Carrier, D. R. (1996). Effect of the respiratory pump on venous return during exercise in iguanas. *Physiologist* **39**, A103.

Forster, H. V. and Pan, L. G. (1988). Breathing during exercise: demands, regulation, limitations. In *Oxygen Transfer from Atmosphere to Tissues*, vol. 227 (ed. N. C. Gonzalez and M. R. Fedde), pp. 257–276. New York: Plenum Press.

Gans, C. and Clark, B. (1976). Studies on ventilation of *Caiman crocodilus* (Crocodilia: Reptilia). *Respir. Physiol.* **26**, 285–301.

Grigg, G. and Johansen, K. (1987). Cardiovascular dynamics in *Crododylus porosus* breathing air and during voluntary aerobic dives. *J. Comp. Physiol.* B **157**, 381–392.

Hernandez, T. and Coulson, R. A. (1980). Anaerobic glycoloysis and repayment of oxygen debt in the alligator. *Comp. Biochem. Physiol.* **67**A, 283–286.

Hicks, J. W. and White, F. N. (1992). Pulmonary gas exchange during intermittent ventilation in the American alligator. *Respir. Physiol.* **88**, 23–36.

Holmes, E. B. (1975). A reconsideration of the phylogeny of the tetrapod heart. *J. Morph.* **147**, 209–228.

Huggins, S. E., Parsons, L. C. and Pena, R. V. (1968). Further study of the spontaneous electrical activity of the brain of *Caiman sclerops*: olfactory lobes. *Physiol. Zool.* **41**, 371–383.

Janicki, J. S., Sheriff, K. K., Robotham, J. L. and Wise, R. A. (1996). Cardiac output during exercise: contributions of the cardiac, circulatory and respiratory systems. In *Handbook of Physiology*, section 12, *Exercise: Regulation and Integration of Multiple Systems* (ed. L. B. Rowell and J. T. Shepherd), pp. 649–704. New York: American Physiological Society.

Lasiewski, R. C. (1963). Oxygen consumption of torpid, resting, active and flying hummingbirds. *Physiol. Zool.* **36**, 122–140.

Lewis, L. Y. and Gatten, R. E., Jr (1985). Aerobic metabolism of American alligators, *Alligator mississippiensis*, under standard conditions and during voluntary activity. *Comp. Biochem. Physiol.* **80**A, 441–447.

Morrison, P., Ryser, F. A. and Dawe, A. R. (1959). Studies on the physiology of the masked shrew *Sorex cinereus*. *Physiol. Zool.* **32**, 256–271.

Naifeh, K. H., Huggins, S. E. and Hoff, H. E. (1970a). The nature of the nonventilatory period in crocodilian respiration. *Respir. Physiol.* **10**, 338–348.

Naifeh, K. H., Huggins, S. E. and Hoff, H. E. (1971). The nature of the nonventilatory period in crocodilian respiration. *Respir. Physiol.* **11**, 178–185.

Naifeh, K. H., Huggins, S. E., Hoff, H. E., Hugg, T. W. and Norton, R. E. (1970b). Respiratory patterns in crocodilian reptiles. *Respir. Physiol.* **9**, 31–42.

Owerkowicz, T., Farmer, C. G., Hicks, J. W. and Brainerd, E. L. (1999). Contribution of gular pumping to lung ventilation in monitor lizards. *Science* **284**, 1661–1663.

Perry, S. F. (1993). Evolution of the lung and its diffusion capacity. In *The Vertebrate Cascade: Adaptations to Environment and Mode of Life* (ed. J. E. Bicudo), pp. 142–153. Boca Raton, FL: CRC Press.

Pough, F. H., Janis, C. M. and Heiser, J. B. (1999). *Vertebrate Life*. Upper Saddle River, NJ: Prentice Hall. 733pp.

Saunders, D. K. and Fedde, R. (1994). Exercise performance of birds. In *Comparative Vertebrate Exercise Physiology: Phyletic Adaptations*, vol. 38B (ed. J. H. Jones), pp. 139–190. San Diego: Academic Press.

Taigen, T. L., Emerson, S. B. and Pough, F. H. (1982). Ecological correlates of anuran exercise physiology. *Oecologia* **52**, 49–56.

Taigen, T. L. and Pough, F. H. (1983). Prey preference, foraging behavior and metabolic characteristics of frogs. *Am. Nat.* **122**, 509–520.

Thomas, S. P. (1981). Ventilation and oxygen extraction in the bat *Pteropus gouldii* during rest and steady flight. *J. Exp. Biol.* **94**, 231–250.

Tucker, V. A. (1968). Respiratory exchange and evaporative water loss in the flying budgerigar. *J. Exp. Biol.* **48**, 67–87.

Walker, A. D. (1970). A revision of the Jurassic reptile *Hallopus victor* (Marsh), with remarks on the classification of crocodiles. *Phil. Trans. R. Soc. Lond.* **257**, 323–371.

Wang, T., Carrier, D. R. and Hicks, J. W. (1997). Ventilation and gas exchange during treadmill exercise in lizards. *J. Exp. Biol.* **200**, 2629–2639.

Weibel, E. R., Taylor, C. R. and Hoppeler, H. (1992). Variation in function and design: Testing symmorphosis in the respiratory system. *Respir. Physiol.* **87**, 325–348.

West, J. B. (1979). *Respiratory Physiology – The Essentials*. Baltimore: Williams & Wilkins. 182pp.

Wright, J. C. (1986). Effects of body temperature, mass and activity on aerobic and anaerobic metabolism in juvenile *Crocodylus porosus*. *Physiol. Zool.* **59**, 505–513.