Metabolic Rate, Q_{10} and Respiratory Quotient (RQ) in Crocodylus porosus, and Some Generalizations about Low RQ in Reptiles

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METABOLIC RATE, Q_{10} AND RESPIRATORY QUOTIENT (RQ) IN CROCODYLUS POROSUS, AND SOME GENERALIZATIONS ABOUT LOW RQ IN REPTILES.[1]

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In juvenile *Crocodylus porosus*, the dependence of oxygen consumption on temperature can be described by the relationship $\log_{10}Q_{O_2} = 0.7221 + 0.0428T$ C. The Q_{10} was found to be 2.68 over the temperature range 20–33 C. No trend was observed between body weight and weight-specific oxygen consumption. At T_a (27–33 C) respiratory quotient (RQ) (measured gasometrically) was invariably low (0.32 — 0.74, mean 0.49, 0.13 SD) in animals from freshwater. The low values of gasometric RQ are attributed to incorporation of carbon dioxide in the urine, forming ammonium bicarbonate. Support for this hypothesis is afforded by the similarity of measured values (0.49) to values predicted by the hypothesis (0.51).

INTRODUCTION

This study was undertaken to describe the effect of temperature on standard metabolic rate and to determine respiratory quotient (RQ) in Australia's Estuarine Crocodile, *Crocodylus porosus*. The paucity of such information for crocodilians has been referred to by Bennett and Dawson (1976) in an excellent review of the relevant literature.

Respiratory quotient values are traditionally regarded as a source of information about the nature of the foodstuff being catabolized, and normal values range from 0.7 for fat to 1.0 for carbohydrate (Schmidt-Nielsen 1975). Values of RQ as low as one-half "normal" values have been reported in reptiles since early days (Hall 1924; Potter and Glass 1931; Benedict 1932; Cook 1949). These low values correspond to the catabolism of no known foodstuff and various explanations for the results have been advanced including hypothetical biochemical events, time lags in changes in the alkaline reserve of the blood, and experimental error. However, Coulson and Hernandez (1959, 1964) suggested that the large amount of ammonium bicarbonate excreted by alligators could be derived from respiratory CO_2, leading to "erroneously" low values for RQ. This was referred to by Smith (1975), who found variable RQ in the alligator (including low values), yet neither Smith nor Coulson and Hernandez referred to the dilemma of low reptilian RQ discussed at length by Benedict (1932). Likewise, Bennett and Dawson (1976) made no reference to Coulson and Hernandez, whose observations suggest that the most likely explanation lies in the formation of ammonium bicarbonate in the urine.

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Low RQ in reptiles will be further discussed in this paper, with reference to Australia's Estuarine Crocodile, *Crocodylus porosus*, which is found in freshwater, estuarine, and marine situations. In freshwater, *C. porosus* excretes mainly ammonia but is uricotelic in the sea (Grigg, in preparation). Coulson and Hernandez (1964) interpreted high levels of bicarbonate in the urine of all alligators (kept in freshwater) as having a buffering function against urinary ammonia. On this basis one would expect low levels of urinary bicarbonate in uricotelic *C. porosus*. This has been confirmed recently in a study of wild-caught *C. porosus* (Grigg, in preparation) where high levels of urinary bicarbonate were found in freshwater (40.5 mmole $1^{-1} \pm 4.4$ SE, no. = 24) compared with values ranging from 0–2.0 mmole 1^{-1} in nine individuals caught in saltwater. If the source of the bicarbonate in urine from freshwater acclimatized *C. porosus* is respiratory CO_2, one would predict low RQ in crocs from freshwater. In saltwater however, higher values of RQ would be expected. Accordingly, RQ was determined in crocodiles with freshwater history and also in the same animals after a period of time in saltwater.

MATERIAL AND METHODS

Animals.—Eleven juvenile *Crocodylus porosus* (180–6,200 g) were used in the study. All were captured from freshwater in the Liverpool River or its tributaries, Mungardobolo and Maradgulidban Creeks, in Arnhem Land, Northern Territory. Following completion of the experimental work, each animal was released at the site of its capture.

Respirometry.—Experimental work was carried out at the Crocodile Research Facility, Maningrida, at the mouth of the Liverpool River on Australia's northern coast. A small insulated air-conditioned room served as a temperature-controlled laboratory.

A simple closed-system constant pressure respirometer was constructed (fig. 1). Respirometer vessels of either 10-cm or 15-cm inside diameter were used, depending upon the size of the crocodile under consideration. Each vessel was a length of PVC drainpipe with one end capped and the other fitted with a threaded lid and O-ring seal for easy introduction or removal of animals. Circulation of air within the system was provided by an aquarists air pump, modified by adding an inlet tube. Air from the respirometer vessel was pumped through a CO_2 scrubber of 0.1 N $Ba(OH)_2$ in a flask, and then via a silica-gel drying bottle to a Servomex Oxygen Analyzer. Confirmation of airflow through both the measurement cell and the bypass of the O_2 analyzer was obtained using GAP flowmeters. To maintain constant pressure during an experiment, as measured with a simple U-tube manometer connected to the vessel, olive oil was pumped from the reservoir into the compensation vessel. This transfer offset the volume changes resulting from absorption of carbon dioxide by $Ba(OH)_2$. Readings of oxygen saturation were made during brief stoppages of airflow. Oxygen consumption was calculated from the measured depletion of oxygen from the gaseous volume of the system.

Carbon dioxide production was determined at the end of each run by titration of duplicate samples of the $Ba(OH)_2$ with 0.1 N HCl using phenolphthalein as an indicator. End points were consistent and sharp and did not differ by more than 2%. In each experiment, either 0.5 liter (larger vessel) or 0.25 liter (smaller vessel) of fresh- or saltwater was added to the respirometer in order to continue the animals' experience of either fresh- or saltwater during the experiment. This

water represented about 3% of the total volume. Calculations showed that any changes in the level of oxygenation of the water during the course of an experiment could have only a negligible effect on the assessment of oxygen consumption. Accordingly, although the volume of the water was subtracted from the total gaseous volume, its contribution as an "oxygen store" was ignored.

Experimental design.—Within 3 days after capture, oxygen consumption and carbon dioxide production of each crocodile were determined at T_a which varied from 27 to 33 C depending on the weather. Because of the small diurnal fluctuations in temperature at this time of the year, and the insulation of the room, T_a within the room stayed essentially constant during the 5–6 h for which any one series of measurements lasted. Before any measurements were made, the animal was given several hours in the respirometer to settle down. After measuremet at the higher temperatures, the crocodile and the room were cooled, usually overnight, to a low temperature (20–22 C) and a second run made on the following day. This allowed determination of Q_{10} and also the effect of temperature on respiratory quotient. Each crocodile was then placed in a holding tank containing seawater whose salinity was boosted to 40‰–45‰ using NaCl. (This exposure to an artificially hypersaline environment was part of another series of experiments to determine the upper limits of salinity at which *C. porosus* can maintain the stability of its plasma osmotic pressure.) Holding

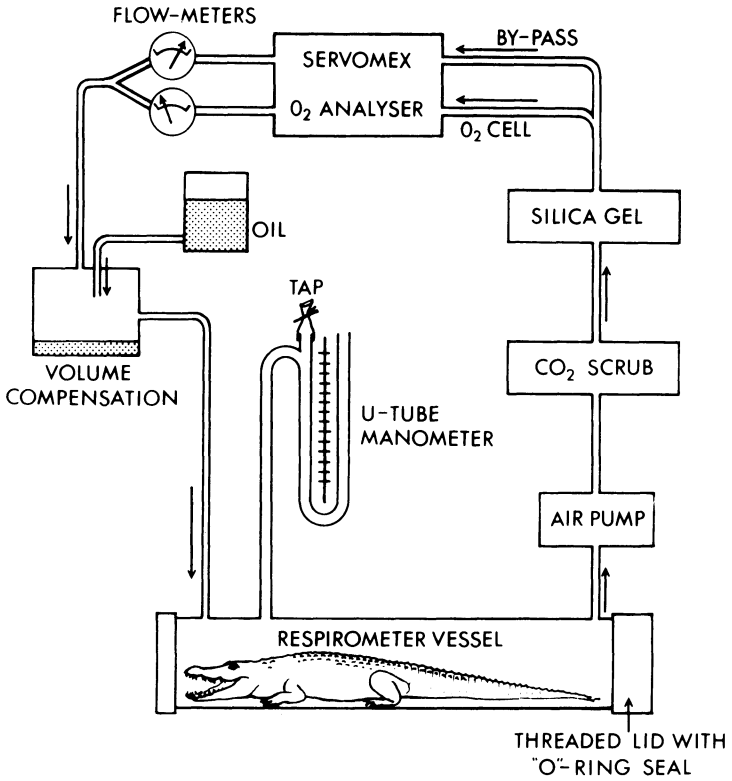

FIG. 1.—Schematic diagram of respirometer used for measurement of oxygen consumption and carbon dioxide production in *Crocodylus porosus*.

tanks were at room temperature. After 2–4 days exposure to the high salinity, respirometry was again carried out at room temperature. Each animal was then released. Analysis of data was performed by standard techniques as described by Snedecor and Cochran (1973).

RESULTS

OXYGEN CONSUMPTION (FIG. 2)

Data gained from animals acclimatized to freshwater were analyzed by multiple-regression techniques to determine effects of temperature and weight on oxygen consumption. The inclusion of weight in the regression did not contribute significantly to explanation of the observed variability. That is to say, the data gave no evidence of a significant relationship between the rate of oxygen consumption and body weight. Accordingly, the second variable was dropped and the data reanalyzed by linear-regression techniques. The line of best fit (fig. 2) is described by the equation $\log_{10}Q_{O_2} = 0.7221 - 0.0428\ T$ C, whose slope was significant ($P < .001$).

The temperature dependence of oxygen consumption by ectotherms is often expressed as Q_{10}; (Schmidt-Nielsen 1975). In the present study a Q_{10} value of 2.68 obtains over the temperature range from 20 to 33 C.

After acclimation to saltwater, mea-

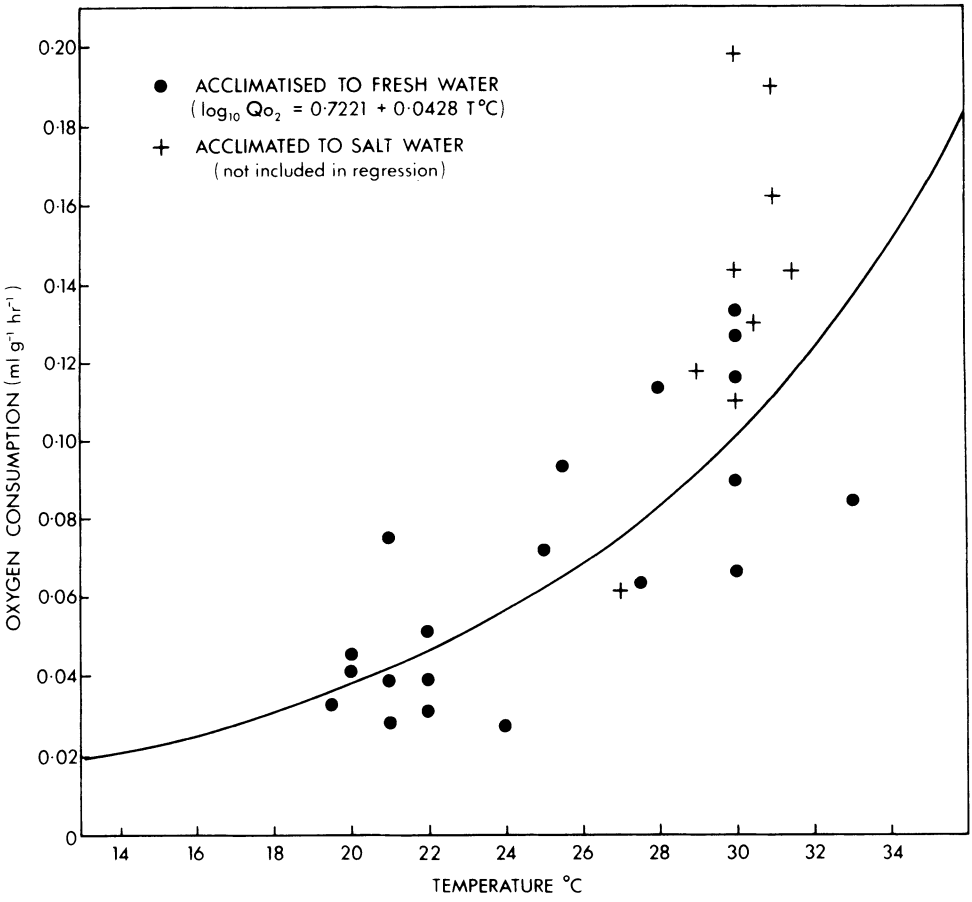

FIG. 2.—The relationship between oxygen consumption and temperature in *Crocodylus porosus*

surements of oxygen consumption were made only at room temperatures. These data are included in fig. 2. Despite a trend toward higher oxygen consumption by animals acclimated to saltwater, the means are not significantly different.

RESPIRATORY QUOTIENT (FIG. 3)

At room temperatures (27–33 C), RQ measured by respirometry of crocodiles living in freshwater was invariably low; mean 0.49 (range 0.32–0.74). Comparisons between animals from freshwater and saltwater showed no apparent differences, and this was confirmed by statistical analysis. The RQ increased at lower temperatures as described by the equation $RQ = 1.098 - 0.0203\ T$ C, whose slope is significant ($P < .001$).

DISCUSSION

OXYGEN CONSUMPTION AND Q_{10}

Fewer than a dozen previous studies have measured oxygen consumption in crocodilians. Bennett and Dawson (1976) have reviewed the studies and presented data which allow comparison with results from *Crocodylus porosus*. Although the results from all studies are quite variable, it can be said that the magnitude of oxygen consumption in *C. porosus* in the present study is similar to that of other crocodilians and, indeed, to other reptilian orders. Coulson and Hernandez (1964) report results from which it is possible to calculate an approximate Q_{10} of 3.4 for *Alligator mississippiensis* between 15 and 30 C. This is a much

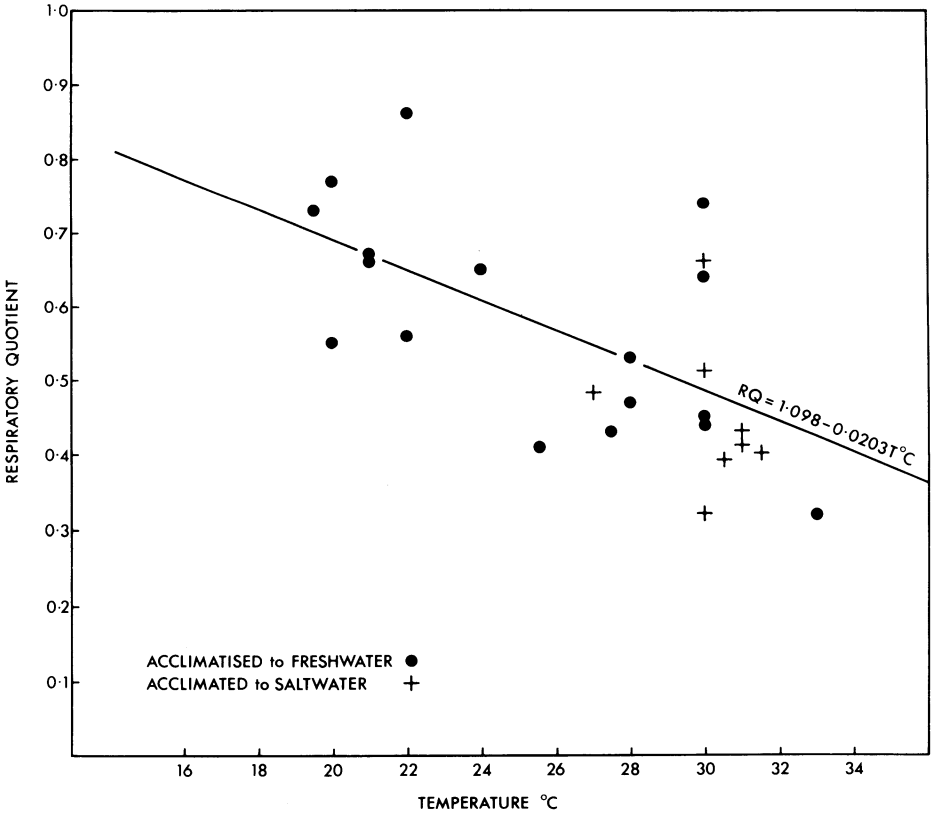

FIG. 3.—The relationship between respiratory quotient and temperature of *Crocodylus porosus*

steeper effect of temperature than that seen in *C. porosus*. The only other data available for comparison of Q_{10} are those of Smith (1975), who heated and cooled alligators while measuring a number of variables including oxygen consumption. He reported a Q_{10} of 3.9 (approximately). Using his regression equation however, I calculated a value of 3.09 for the same temperature range (15–35 C). The value reported for Q_{10} in *C. porosus*, 2.68, is consistent with what one would expect for a reptile (Bennett and Dawson 1976). The failure of weight to contribute significantly to explanation of the total variability was surprising, especially in view of the large size range of the animals. Perhaps age-related factors offset the expected relationship between body weight and oxygen consumption per kilogram.

RESPIRATORY QUOTIENT

The RQ in fresh- and saltwater.—At ambient temperatures *C. porosus* was found to have a low RQ in freshwater, as proposed on the basis of earlier observations of high concentrations of ammonium bicarbonate in the urine (see Introduction). However, low values of RQ persisted after exposure of the animals to saltwater, even though solid urate pellets were being produced and the animals were clearly uricotelic. Low RQ in saltwater was unexpected in terms of the initial hypothesis, but two factors contribute to an explanation of the observations. In the first place, subsequent analyses of urine samples taken from four of the crocodiles kept in saltwater in the laboratory showed variable, but in three cases substantial, amounts of bicarbonate present (2.0, 6.3, 30.0, and 40.0 mmol l^{-1}). The experimental exposure of animals to saltwater for a few days appears, therefore, to have been an ineffective way of mimicking the field situation seen in crocodiles fresh caught from saltwater where bicarbonate is present in the urine only in very small amounts. Second, Roberts (1968) has pointed out that uricotelism leads to reduction of RQ values from 0.8 for protein catabolism to about 0.7. It seems likely that, had animals with a natural saltwater history been available for measurement, RQ would have been higher than for freshwater history animals.

Higher RQ at low temperatures.—Higher values of RQ were observed at low temperatures (fig. 3), after the crocodiles had been exposed to the lower temperature overnight. Shift to a lower temperature should give rise to a transient decrease in RQ as carbon dioxide is sequestered in the blood to make up a larger alkaline reserve at low temperatures (Bennett and Dawson 1976). However, calculations suggest that such a decrease would last no more than a couple of hours. The increase in RQ at low temperatures may reflect a decrease in the relative amount of protein among substrates being catabolized, concomitant with the overall reduction in metabolic rate at low temperature. This may result in a lesser production of ammonium bicarbonate and, hence, a higher RQ.

Comparison between measured and predicted values.—If carbon dioxide which would otherwise have been expired as a gas is excreted by ammonotelic crocodiles as bicarbonate, thereby buffering ammonia in the urine, one can predict the RQ for the whole animal as follows:

Consider a 1,000-g *C. porosus* at 25 C. From figure 1, it can be calculated that oxygen consumption will be 1,440 ml per day. In a carnivore, one would expect protein to be a major substrate for catab-

olism, giving a tissue RQ close to 0.8 and a daily CO_2 production of 1,152 ml. Now, 960 ml O_2 oxidize 1 g protein (dry weight) and produce approximately 0.16 g nitrogen (Schmidt-Nielsen 1975). Therefore consumption of 1,440 ml O_2 is equivalent to the production of 0.24 g nitrogen which, as ammonium ion, would require 0.0171 mol of carbon dioxide as a source for bicarbonate. This represents 418 ml of gaseous CO_2 at 25 C. So we can say that of 1,152 ml CO_2 produced each day by the tissues, 418 ml contribute to urinary bicarbonate. This leads to an overall gaseous exchange for the animal of 1,440 ml O_2 uptake and 734 ml gaseous CO_2 released, i.e., RQ = 0.51.

This predicted value agrees very favorably with the measured values.

"Tissue RQ" and "apparent RQ."—Understanding of the significance of low RQ in reptiles springs from Coulson and Hernandez (1964), yet their reference to it also gives an interesting insight into the different significance attached to RQ by biochemists compared with comparative physiologists. On page 10 of their remarkable book they say, "Fortunately, technical difficulties . . . prevented attempts to determine the respiratory quotient of the alligator. At this time we were unaware of the role played by the kidney in the excretion of very great amounts of CO_2 in the form of NH_4HCO_3, and by sheer luck were spared the embarrassment of the publication of erroneous data." Their clear implication is that a measurement of RQ is in error unless it reflects in good measure the gas exchange accompanying tissue metabolism. It may be useful to use the term "apparent RQ" for measurements of RQ (made gasometrically) of the whole animal. Values of apparent RQ which are different from an expectation based on tissue metabolism may indicate events of interest to the physiologist. In the present case, low values of apparent RQ result from neither experimental error nor metabolic trickery but from high values of urinary bicarbonate. This may well be the case in other reptiles and in ammonotelic animals whose major route of nitrogen excretion is urinary.

LITERATURE CITED

BENEDICT, F. G. 1932. The physiology of large reptiles. Carnegie Inst. Washington Pub. 425.

BENNETT, A. F., and W. R. DAWSON. 1976. Metabolism. Pages 127–223 *in* C. GANS and W. R. DAWSON, eds. Biology of the reptilia. Vol. 5. Academic Press, New York.

COOK, S. F. 1949. Respiratory metabolism of certain reptiles and amphibia. Univ. Calif. Pub. Zool. **53**:367–376.

COULSON, R. A., and T. HERNANDEZ. 1959. Source and function of urinary ammonia in the alligator. Amer. J. Physiol. **197**:873–879.

———. 1964. Biochemistry of the alligator: a study of metabolism in slow motion. Louisiana State University Press, Baton Rouge.

HALL, F. G., 1924. The respiratory exchange in turtles. J. Metabolism Res. **6**:393–401.

POTTER, G. E., and H. B. GLASS. 1931. A study of respiration in hibernating horned lizards, *Phrynosoma cornutum*. Copeia **1931**:128–131.

ROBERTS, L. A. 1968. Oxygen consumption in the desert lizard *Uta stansburiana*. Ecology **49**:809–819.

SCHMIDT-NIELSEN, K. 1975. Animal physiology—adaptation and environment. Cambridge University Press, London.

SMITH, E. N. 1975. Oxygen consumption ventilation and oxygen pulse of the American alligator during heating and cooling. Physiol. Zool. **48**:326–337.

SNEDECOR, G. W., and W. G. COCHRAN. 1973. Statistical Methods. Iowa State University Press. Ames.