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Diving through the thermal window: implications for a warming world

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Population decline and a shift in the geographical distribution of some ectothermic animals have been attributed to climatic warming. Here, we show that rises in water temperature of a few degrees, while within the thermal window for locomotor performance, may be detrimental to diving behaviour in air-breathing ectotherms (turtles, crocodilians, marine iguanas, amphibians, snakes and lizards). Submergence times and internal and external body temperature were remotely recorded from freshwater crocodiles (*Crocodylus johnstoni*) while they free-ranged throughout their natural habitat in summer and winter. During summer, the crocodiles' mean body temperature was 5.2 ± 0.1°C higher than in winter and the largest proportion of total dive time was composed of dive durations approximately 15 min less than in winter. Diving beyond 40 min during summer required the crocodiles to exponentially increase the time they spent on the surface after the dive, presumably to clear anaerobic debt. The relationship was not as significant in winter, even though a greater proportion of dives were of a longer duration, suggesting that diving lactate threshold (DLT) was reduced in summer compared with winter. Additional evidence for a reduced DLT in summer was derived from the stronger influence body mass exerted upon dive duration, compared to winter. The results demonstrate that the higher summer body temperature increased oxygen demand during the dive, implying that thermal acclimatization of the diving metabolic rate was inadequate. If the study findings are common among air-breathing diving ectotherms, then long-term warming of the aquatic environment may be detrimental to behavioural function and survivorship.

Keywords: crocodilian; ectotherm; acclimatization; temperature; aerobic dive limit

1. INTRODUCTION

An ectotherm is by definition an animal whose body temperature varies with that of the ambient environmental temperature. Apart from one or two exceptions, these include all the fishes, amphibians, reptiles and invertebrates. The overbearing influence of temperature on the rate of metabolic processes means that for these animals ambient environmental temperature strongly influences ecological function (Portner & Farrell 2008). This close association means that acute and chronic shifts in ambient thermal conditions may be detrimental to an individual's survival, and numerous cases have recently been reported of population decline and a shift in the geographical distribution of some ectotherms as a direct result of climatic warming events (Alford *et al.* 2007; Kutschera *et al.* 2007; Portner & Knust 2007; Farrell *et al.* 2008; McMenamin *et al.* 2008; Takasuka *et al.* 2008).

The structural and kinetic coordination of molecular, cellular and systemic processes results in animals optimizing performance within a limited thermal window (Portner & Farrell 2008). Beyond the upper and lower limits of this window, decrements in locomotion, growth, competitiveness, reproduction and immune competence will occur (Portner 2002). Confronted with a thermally generated reduction in performance, an organism may use behavioural and physiological strategies to mitigate negative effects. Behavioural strategies typically result in an avoidance of extreme ambient environmental temperatures and a selection of favourable thermal niches (Daly *et al.* 2008). If the animal cannot escape adverse ambient temperatures on the seasonal or longer-term time scale, then a reshaping of the thermal window may occur (Wilson & Franklin 1999; Franklin & Seebacher 2009). Knowing the capacity by which an organism can behaviourally compensate or physiologically acclimatize to long-term shifts in ambient environmental temperature may enable better prediction of its ecological response, and thus the long-term consequences of climatic change for the population.

Air-breathing diving animals provide a good model for examining the interplay between physiology and ecological function because diving behaviour is directly determined by the amount of stored oxygen and the rate it is used (Kooyman *et al.* 1980; Costa *et al.* 2004). The limitation by which animals can routinely dive without incurring an oxygen debt is defined by the diving lactate threshold (DLT; Butler 2006). Staying submerged beyond the limits of the DLT rapidly builds up

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oxygen debt and results in exponential lengthening of the post-dive surface interval (PDSI) or, depending on the extent, a suspension of diving until normal lactate levels are re-established (Kooyman *et al.* 1980; Costa *et al.* 2004).

The close association between ambient temperature and body temperature in ectothermic animals means that at higher environmental temperatures, metabolic rate is proportionally increased (Q_{10} effect; Pough 1976; Jackson 2007). This causes a reduction in aerobic dive limit as a consequence of O_2 being used more rapidly, and a reduction in available O_2 stores (Herbert & Jackson 1985; Fuster *et al.* 1997; Prassack *et al.* 2001; Priest & Franklin 2002; Gordos *et al.* 2003*a*,*b*; Seebacher *et al.* 2005; Jackson 2007). Subsequently, a diving ectotherm faced with a higher body temperature would be required to reduce dive duration to maintain the dive within aerobic limits or, if dive duration remained constant, increase the duration of the PDSI to clear the anaerobic debt (lactate). Either of these modifications would reduce the amount of time the animal could remain submerged during a bout of diving activity.

Present and projected rises in the temperature of some marine and freshwater environments (Intergovernmental Panel on Climate Change 2007) would increase the body temperature of inhabiting ectothermic air-breathing diving organisms (marine and freshwater turtles, crocodilians, marine iguanas, sea and freshwater snakes), and may impinge on their diving performance. If this occurred, the animal's ability to undertake diving-related ecological tasks could be impeded, survivorship reduced and a shift in the geographical distribution of the species may occur. Although it is known that acute rises in temperature significantly reduce dive performance in ectotherms (Priest & Franklin 2002; Clark *et al.* 2008), it is presently unclear how the diving performance of ectotherms responds to chronic rises in environmental temperature. Do ectotherms undergo adequate physiological acclimatization of the diving metabolic rate or is diving performance diminished?

This is a difficult question to answer empirically because numerous ecological factors influence the diving behaviour of an animal in the wild, while, in captivity, the environmental cues and stimulus for defining diving behavioural strategies are not present. In this study, we remotely monitored the diving behaviour, internal body temperature and the preferred microhabitat temperature of juvenile freshwater crocodiles (*Crocodylus johnstoni*) in the wild. The freshwater crocodile was a suitable ectothermic air-breathing diver to study; firstly, because it lives in an environment where it is exposed to a seasonal shift in environmental temperatures (approx. 8°C), but winter temperatures are not sufficiently low to induce hypometabolism (Seebacher & Grigg 1997; Seebacher *et al.* 2005); and, secondly, because they rest/sleep for prolonged periods on the river or waterhole substratum, surfacing to breathe when they approach their aerobic dive limit (Campbell *et al.* 2010). Thus, the effect of a long-term rise in temperature upon diving performance could be assessed in a free-diving ectotherm while minimizing physiological (activity, feeding) and ecological interference (predators, prey, conspecific interactions).

2. MATERIAL AND METHODS

(a) *Animals and surgery*

Freshwater crocodiles (*C. johnstoni*) were captured from a permanent waterhole (approx. 150 m in length, by 20 m wide, and between 1.5 and 3.0 m depth) in Lakefield National Park, Queensland, Australia (15.155840° S, 144.251992° E). The studies were undertaken in mid-June ($n = 10$) and late November 2007 ($n = 10$). Following capture by a set net, the crocodiles were sedated with 2 mg kg^{-1} body mass of Alfaxan CD-RTU (10 mg ml^{-1}; Jurox, New Zealand), and a local anaesthetic (2–4 ml lignocaine, 20 mg ml^{-1} with adrenaline 1 part per 100 000; Troy Laboratories, Australia) was injected into the immediate area of tag attachment or implantation. All crocodiles were responsive to touch and auto-moving 1–2 h after surgery. They were released at the capture location 24 h post-surgery.

Throughout the duration of the study, data-loggers (iButton Thermocron, Dallas Semiconductor, Dallas, TX, USA) recorded air and water temperature at 0.15 (subsurface) and 1.5 m (on the bottom of the waterhole) depth every 20 min, within a shaded area of the waterhole.

(b) *Remote monitoring of depth*

The depth at the crocodiles dorsal surface was logged every 9 s using a time–depth–temperature recorder (TDTR; cylindrical dimensions = 60 mm L × 21 mm D; 15 g; Star-Oddi, Iceland) mounted on the nuchal shield. The device was attached using a plastic-coated multi-strand stainless steel wire to 4 × 1 mm holes drilled transversally through the raised dorsal scutes. Before deployment, all TDTRs were calibrated in the laboratory. Depth and temperature sensors were accurate to a resolution of 0.04 m and 0.1°C, respectively.

(c) *Remote monitoring of temperature*

The micro-niche temperature of each crocodile was recorded every 20 min by the TDTR (see above) mounted on the nuchal shield. The body temperature was measured simultaneously for each TDTR recording by temperature-sensitive radio transmitters (cylindrical dimensions = 40 × 22 mm; 15 g; Sirtrack, NZ) surgically implanted into the peritoneal cavity. A 4 cm lateral incision was made dorsal of the left hind-leg. The muscle layers were teased apart by a blunt dissection and the tag placed in a pocket under the abdominal muscle layer but external to the peritoneal cavity. The muscle layer was closed with two interrupted sutures (3-0 Ethicon cat gut), and the wound closed with six to eight interrupted sutures. Aseptic conditions were maintained throughout surgery. The transmitters emitted a radio pulse at a unique frequency for each animal. The pulse rate from each transmitter was temperature-dependent. The relationship between pulse rate and temperature was calibrated for each transmitter against a certified thermometer prior to implantation with an accuracy less than 0.3°C. During the study, the time elapsed for 10 consecutive pulses was used to calculate temperature.

To record the body temperature from free-diving crocodiles, a large four-prong radio antenna (Titley Electronics, Brisbane, Australia) was positioned at 10 m height above the waterhole. A receiving station was situated 110 m from the waterhole, from which the radio signal could be monitored without disturbance to the crocodiles. A scanning radio-receiver (Titley Electronics) continually recorded the received signals and scanned the frequency for each transmitter every 60 s; subsequently each transmitter was scanned every 10 min. The audio output signal from the radio-receiver was directed to a data acquisition system (Powerlab, ADInstruments, Australia) that digitized the audio signal at a sample rate of 100 Hz. The digital trace was recorded and stored on a laptop computer (Toshiba) running CHART 5.1 software (ADInstruments).

(d) *Statistical analysis*

This study used a combination of general additive mixed models (GAMMs) and general linear mixed models (GLMMs) to describe patterns in diving behaviour and thermoregulation in *C. johnstoni*. General additive models (GAMs) allow for nonlinear relationships between the response variable and multiple explanatory variables (Wood 2004). Mixed models extend this framework, providing estimates of the influence on the mean as well as the influence of random effects on the variance, thereby accounting for the non-independence of errors resulting from repeated measures from the same individuals (Pinheiro *et al.* 2009). Data from temperature sensors were analysed using a GAMM with body temperature as the dependent variable, season as a factor (summer and winter) and crocodile ID as a random effect. A single measure of 'environmental temperature' was derived from a principal components analysis using the correlation between deep water, subsurface water and micro-niche temperature to produce a single 'environmental temperature' covariate. Principal component 1 (PC1) accounted for 95 per cent of the variance and was selected as an environmental temperature index (factor loadings for micro-niche temperature, surface water temperature and deep water temperature were 0.58, 0.70 and 0.41, respectively). The relationship between Tb and each of the environmental temperature measurements was determined for each hour over the diel cycle using an ANOVA with a Bonferroni correction for multiple comparisons.

To extract the dive events within the depth recordings, a custom-written program (M. A. Gordos) in Visual Basic (Microsoft EXCEL, 2003) was used. A dive threshold (0.3 m) was set, upon which a dive event would be scored. From these scored dives, variables were calculated on an hourly basis for each of the 15 day study periods. The presence of harmonic oscillations in diving repertoire was determined statistically for each crocodile using power spectral analysis. The mean dive duration, maximum dive depth, number of dives and proportion of time submerged were calculated on an hourly basis and were then subjected to the Lomb–Scargle periodogram procedure before undergoing a discrete Fourier transform (MATLAB R2008a). The results were outputted graphically, and, if rhythmicity occurred in the data, a peak was shown at the designated frequency and a significance value (PS) given. Peaks with a PS > 0.05 were deemed to show a significant sigmoid oscillation in the dataset. To determine the influence of covariates upon dive duration, a second GAMM was employed. Dive duration was log-transformed prior to analysis and set as the dependent variable. Body mass and time of dive commencement were set as covariates, season as a factor and crocodile ID as a random effect. A GLMM was used to examine the association between dive duration and the PDSI, with a log-transformed PDSI as the dependent variable, dive duration and body mass as covariates, season as a factor and crocodile ID as a random effect. The effect of season upon mean dive duration, dive number, proportion of each hour submerged and maximum dive depth was determined for each hour over the diel cycle using an ANOVA with a Bonferroni correction for multiple comparisons. The Mann–Whitney *U*-test and the Kolmogorov–Smirnoff test were used to make seasonal comparisons upon median and distribution data, respectively. Welch's *t*-test assuming unequal variances was used to examine mean differences in non-diving parameters between the two sampling periods. Spearman's rank-order correlation was used to examine the relationship between dive duration and the PDSI for each animal. All modelling was undertaken using the R statistical software package (R Development Core Team 2005). GLMMs and GAMMs were carried out with the NLME (Pinheiro *et al.* 2009) and MGCV (Wood 2004) packages, respectively. ANOVAs were undertaken in STATGRAPHICS v. 5.1, and an effect was deemed significant if $p < 0.05$.

Table 1. Environmental and internal body temperature (Tb) in *C. johnstoni* (winter, $n = 9$; summer $n = 6$; mean ± s.e.m.). All values in °C. Asterisks denote a significance difference between the seasons, $p < 0.05$.

	winter	summer
air	18.5 ± 4.2*	27.7 ± 4.2*
surrounding rock	18.9 ± 6.4*	31.3 ± 2.9*
surface water	21.3 ± 4.4*	29.1 ± 2.5*
deep water	23.6 ± 0.2*	28.4 ± 1.3*
daily micro-niche temp	23.9 ± 0.02	29.0 ± 0.02
daily mean Tb	23.9 ± 0.02*	29.1 ± 0.15*
daily min Tb	24.2 ± 0.12	28.0 ± 0.53
daily max Tb	27.2 ± 0.51*	32.2 ± 0.27*

3. RESULTS

(a) *Temperature*

The average air temperature was 9.2°C warmer in summer compared with winter (table 1). Surface water temperature showed a seasonal shift of 7.8°C, while deep water temperature showed a seasonal variation of only 4.8°C. This resulted in a different thermal stratification in the water column between the seasons (figure 1). In winter, surface waters, warmed throughout the morning, were 1–2°C warmer than deep water by 14.00, cooled rapidly after 15.00 and by 04.00 were 4.2°C cooler than the deep water. In summer, surface waters were 3.4°C warmer than deep water at 14.00 and never cooled to a temperature below that of the deep water. The mean daily micro-niche temperature of the crocodiles was 5.1°C warmer in summer than in winter (table 1). Micro-niche temperature followed a similar daily profile to surface water temperature for seasons, increasing rapidly during the morning and decreasing throughout the late afternoon. In winter, however, micro-niche temperature remained elevated over surface water temperature throughout the night, and was comparable to the deep water temperature (figure 1*a*). In summer, micro-niche temperature remained similar to surface water temperature throughout the night, but was significantly cooler than surface water temperature between 14.00 and 16.00 (figure 1*b*).

Using the principal component for environmental temperature, the GAMM indicated that Tb was affected by environmental temperature equally across summer and winter (REML: $F_1 = 0.63$, $p = 0.425$; figure 2). The relationship between the seasons was linear, but a

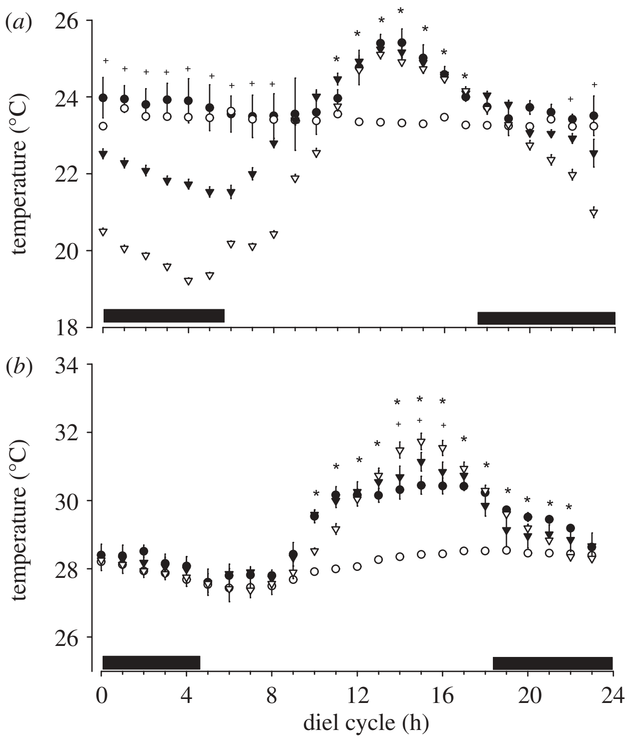

Figure 1. The relationship between the body temperature (Tb) of *C. johnstoni* (black circles; mean ± s.e.; $n = 8$) with micro-niche (black triangles; mean ± s.e.; $n = 8$), water subsurface (white triangles; mean ± s.e.; $n = 45$) and deep water (white circles; mean ± s.e.; $n = 45$) temperature for each hour over the diel cycle in (*a*) winter and (*b*) summer. Plus symbols denote hours when Tb was significantly different from surface water temperature, and asterisks denote hours when Tb was significantly different from deep water temperature.

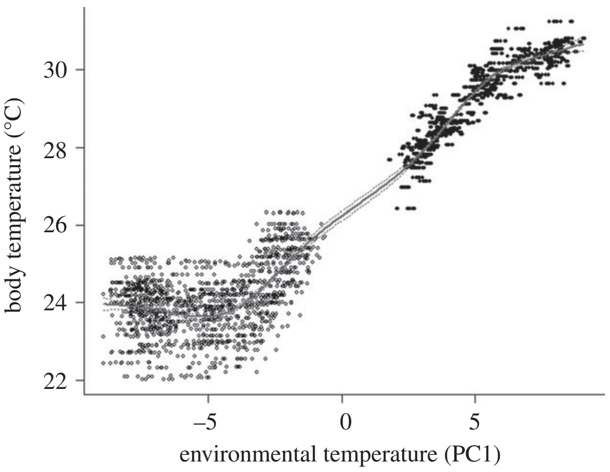

Figure 2. The best-fitting GAM model representing the relationship between the body temperature of *C. johnstoni* and environmental temperature (PC1) across both winter (white circles) and summer (black circles). Environmental temperature was derived from a principal component analysis (PCA) of the micro-niche temperature, water surface temperature and deep water temperature (REML: $F_1 = 0.63$, $p = 0.425$).

Table 2. Diving behaviour recorded in free-ranging *C. johnstoni* (winter, $n = 9$; summer = 6, mean ± s.e.m., median ± s.d.). Asterisks denotes a significance difference between the seasons, $p < 0.05$.

season	winter	summer
median dive duration (min)	14.70 ± 0.26*	12.51 ± 0.23*
max dive duration (min)	77.01 ± 10.34*	48.77 ± 3.62*
no. dives per crocodile per day	47.94 ± 5.59*	66.16 ± 5.51*
time submerged (%)	38.52 ± 3.27	43.04 ± 4.31
post-dive surface interval (min)	5.22 ± 0.86	4.92 ± 0.95
dive depth (cm)	114.23 ± 18.25*	92.31 ± 15.43*

nonlinear logistic function best fitted the data (REML: $F_{8.34} = 1862$, $p < 0.01$). This occurred because of dissociation between Tb and environmental temperature from linearity within each season and not between seasons. The within-season departure from linearity occurs because Tb does not equal the high surface water temperatures during summer afternoons or the low surface water temperatures during winter nights (figure 1). During these periods, Tb was more similar to micro-niche temperature than surface water temperature ($p < 0.05$). This modification of Tb by micro-niche selection resulted in Tb varying by approximately 2°C over the diel cycle in both summer and winter, despite the diel change in surface water temperature varying by greater than 6°C. It also helped to explain the reduced seasonal difference in Tb (5.2°C) compared with the seasonal variation in surface water temperature (7.8°C).

(b) *Diving behaviour*

In total, 6472 dives were recorded in winter, compared with 5955 dives in summer. However, data were successfully retrieved from more crocodiles in winter, and the mean number of dives exhibited per day was 38 per cent greater in summer (d.f. = 6 and 9, $W = 0.32$, $p < 0.05$). The median and maximum dive durations were significantly shorter in summer compared with winter (d.f. = 6 and 9, $W = 1.52$, $p < 0.05$; table 2), and the proportion of each day that the crocodiles were submerged did not vary between the seasons (d.f. = 6 and 9, $W = 7.52$, $p = 0.76$). In both seasons, a large majority of dives were less than 20 min duration; during winter, however, a substantially greater proportion of time was spent on dives more than 40 min (figure 3). In summer, the crocodiles undertook 0.83 ± 0.07 (mean ± s.e., $n = 6$) dives of more than 50 min in duration, compared with 12.6 ± 0.34 (mean ± s.e., $n = 9$) in winter.

The mean body mass of the crocodiles recorded in winter and summer was 7.3 ± 1.2 and 6.8 ± 2.1 kg, respectively. There was no significant difference in mean body mass between the two sample periods ($t_{9,6} = 1.65$; $p = 0.12$); however, the relationship between body mass and dive duration was significantly different between the seasons (REML: $F_1 = 30.36$, $p < 0.01$). In summer, there was a positive nonlinear effect between dive duration and body mass, while in winter, body mass had little effect on dive duration (figure 4*a*).

In both seasons, dive duration increased at first light, approximately 1 h earlier in summer, illustrating an association between diving behaviour and photoperiod. A significant circadian rhythm was present in the duration

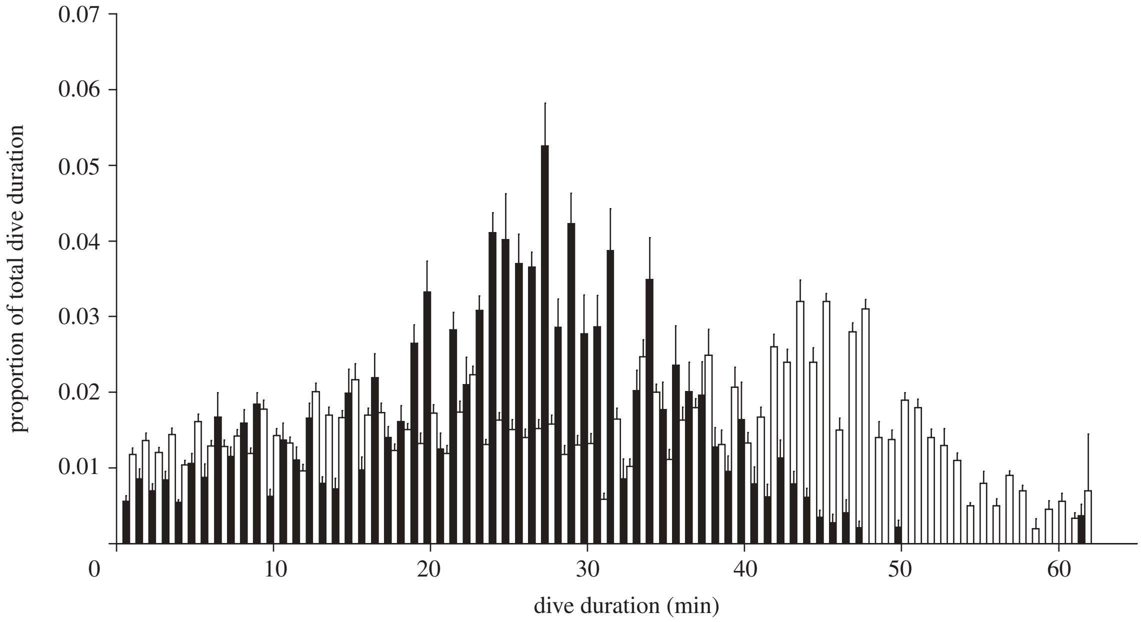

Figure 3. Dive duration of *C. johnstoni* (mean + s.e.) recorded in winter ($n = 9$; white bars) and summer ($n = 6$; black bars) as a proportion of the total time within each duration bin. Dive duration bins represent 0.8 min increments.

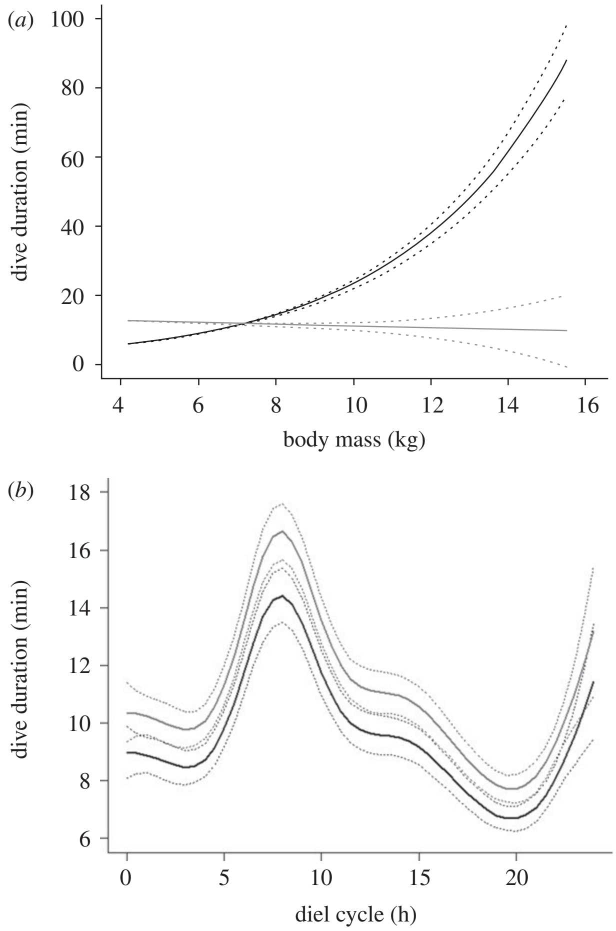

Figure 4. The best-fitting GAM model representing the relationship between dive duration of *C. johnstoni* with (*a*) body mass (REML: $F_1 = 30.36$, $p < 0.01$) and (*b*) diel cycle (REML: $F_{8.02} = 49.80$, $p < 0.01$). The influence of winter (grey line) and summer (black line) upon this relationship is shown.

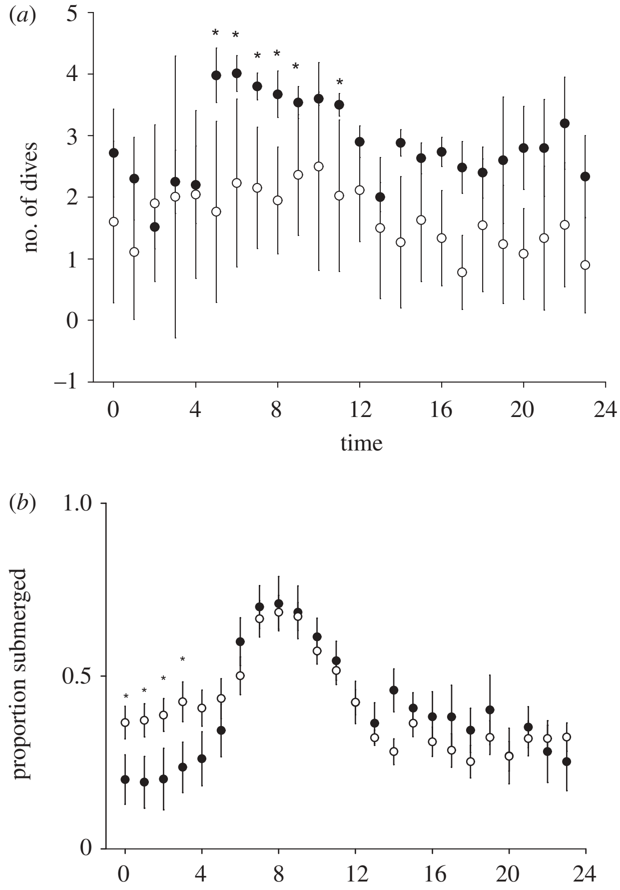

Figure 5. (*a*) The number of dives and (*b*) the proportion of time submerged by *C. johnstoni* over the diel cycle (winter, white circles, $n = 9$; summer, black circles, $n = 8$). Asterisk signifies hours that showed a significant difference between seasons.

of the dives. The period of the circadian rhythm was 22.2 and 26.4 h (PS < 0.05). The period of the sine wave was similar for both seasons, and the relationship between dive duration and time of day was best explained by a nonlinear GAMM (REML: $F_{8.02} = 49.80$, $p < 0.01$). However, dive duration during winter months was significantly greater at each period throughout the diel cycle

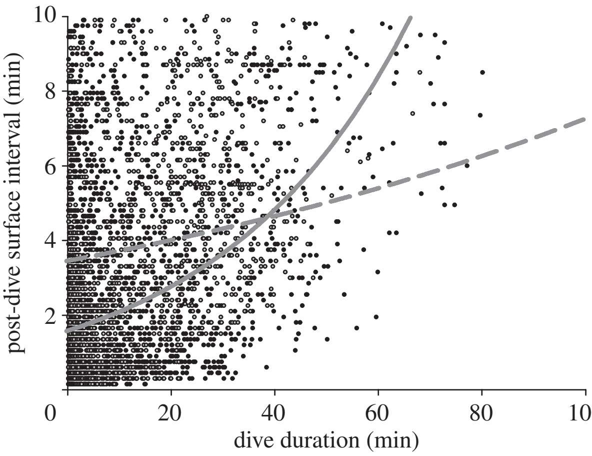

Figure 6. Dive duration of *C. johnstoni* plotted against the PDSI during winter ($n = 9$, white circles) and summer (summer, n=6, black circles). The best-fitting GLM model is shown to represent the relationship (REML, $\chi_1^2 = 5.48$, $p = 0.019$) during winter (dashed line) and summer (solid line).

(REML: $F_1 = 24.41$, $p < 0.01$; figure 4*b*). During this morning period when dive duration was at its daily maximum, the crocodiles engaged in a significantly greater number of dives in summer ($W = 2.3 \times 10^5$, $p < 0.01$; figure 5*a*). This resulted in no significant difference ($W = 2.8$, $p = 0.078$) between the seasons in the proportion of time they were submerged during this morning bout of extended diving (figure 5*b*).

The median duration of the PDSI was not significantly different between the seasons ($W = 4.64$, $p = 0.99$), nor was there a significant difference in the distribution of data (K–S = 0.36, $p = 0.78$). This occurred even though median dive duration was significantly greater during winter months (table 2). When analysed individually, all *C. johnstoni* showed a significant positive correlation between the duration of the dive and the duration of the PDSI (Spearman's ranked-order correlation, $0.11 < r_s < 0.35$, $p < 0.05$); longer dives resulted in the crocodiles extending the duration of the PDSI. The interaction between season and dive duration was highly significant in predicting PDSI in the GLM model (REML: $\chi_1^2 = 5.48$, $p = 0.019$; figure 6). The relationship between dive duration and PDSI showed significant departure between the seasons when dive duration extended beyond approximately 40 min. Crocodiles undertaking dives progressively greater than 40 min in duration were required to exponentially increase the length of the PDSI during summer, while in winter a more gradual increase in PDSI was observed.

4. DISCUSSION

Many species of ectothermic tetrapods are aquatic or semiaquatic and participate in diving to perform ecological tasks. In this study, we observed freshwater crocodiles (*C. johnstoni*) diving in their natural habitat during summer and winter months, and observed mean body temperature (Tb) to be 5.2 ± 0.1°C higher in summer and average dive duration significantly reduced compared with winter. The interaction between dive duration with body mass and PDSI strongly suggests that the reason for the reduced dive performance in summer was because of a higher rate of oxygen consumption during the dive. Because the crocodiles were inactive during the majority of these extended dives (Campbell *et al.* 2010), diving metabolic rate must have been greater. This result is similar to the effects of acute thermal exposure on diving in air-breathing ectotherms (Fuster *et al.* 1997; Priest & Franklin 2002; Jackson 2007), and implies that inadequate thermal acclimatization of the diving metabolic rate occurred between summer and winter.

This finding appears odd as numerous examples exist of ectotherms altering enzyme activity, cardiovascular function and metabolism in direct response to chronic changes in environmental temperature (Zari 1999; Hicks & Farrell 2000; Birkedal & Gesser 2004; Seebacher 2005; Seebacher *et al.* 2009), and crocodilians show significant changes in the activities of regulatory metabolic enzymes after only a few weeks of thermal acclimatization (Glanville & Seebacher 2006; Seebacher *et al.* 2009). Most field studies on the thermal biology of reptiles have however focused on their acclimatization to cold temperatures, and ectotherms from temperate regions that use hypometabolic strategies are often the study species.

In this study, behavioural rather than physiological acclimatization appeared to be the more significant seasonal response to temperature. During summer, the duration of the dives was reduced and a greater number of dives were undertaken during periods of the day when the crocodiles spent a high proportion of their time submerged. All *C. johnstoni* showed a significant positive correlation between dive duration and the PDSI in summer; however, diving beyond 40 min required an exponential lengthening of the PDSI. This dive and PDSI pattern is typical of an air-breathing animal diving beyond its DLT, as it clears the oxygen debt built up during each dive (Kooyman *et al.* 1980; Costa *et al.* 2004). The fact that the lengthening of the PDSI did not occur to the same extent during winter suggests that during these periods when Tb was lower, their DLT was extended. The summertime reduction in DLT would arise from a higher rate of oxygen consumption during the dive, as a direct result of a thermally induced higher spontaneous metabolic rate.

In a previous study, we found that the relationship between body mass and dive duration was a poor predictor of dive duration in *C. johnstoni*, and although DLT did scale allometrically with body mass, ecological factors exerted a greater influence on dive duration than oxygen reserves (Campbell *et al.* 2010). In this previous study, however, diving behaviour was studied only during winter months. The present study agrees with the previous findings for crocodiles examined during winter months, but when crocodiles are examined during summer months, dive duration does scale with body mass. This finding supports the theory that the crocodiles' oxygen reserves were significantly more influential in controlling dive duration during summer than during winter, and, because dives were on average shorter during summer, we hypothesize that diving metabolic rate was greater.

It is difficult to assess and quantify the ecological ramifications for *C. johnstoni* as a direct consequence of a reduced dive performance during summer months. We can deduce that all crocodiles compensated for the restricted dive performance by undertaking a greater number of dives during periods when a high level of submergence occurred. Chronic exposure to high ambient temperatures may be more detrimental to hatchlings owing to their higher mass-specific metabolic rates. The crocodiles used in this study were roughly 7 kg; extrapolation of DLT for a 70 g hatchling crocodile (using the allometric scaling exponent for aerobic dive duration in *C. porosus*; Wright & Kirshner 1987) shows that for the rise in Tb observed in this study, the DLT of a hatchling freshwater crocodiles would be reduced from 11.36 to 7.1 min and diving beyond DLT would result in the crocodile needing to spend prolonged periods at the water surface to clear anaerobic debt. Further increases in ambient temperature would decrease DLT and the hatchlings would be required to spend greater periods of time at the water surface and vulnerable to predation from aerial and surface-feeding predators. Additionally, as metabolic rate rises with ambient temperatures, the energetic requirements of the crocodile would also increase. For an air-breathing animal that needs to dive to collect food, even slight restrictions on DLT may be detrimental to their foraging strategy, requiring them to push physiological limits and undertake a larger proportion of energetically costly anaerobic dives. A negative feedback loop can be envisaged, where the animal must engage in high-energy-cost foraging behaviour to satisfy growing metabolic demands.

All organisms exhibit a thermal window for optimal locomotor and metabolic performance. Living outside the limits of this window will reduce fitness, but within window boundaries, a broad plateau of thermal independence exists (Portner & Farrell 2008). The thermal constraints upon diving performance in *C. johnstoni* occurred under ambient temperatures, and transpired even though Tb fell well within their thermal window for locomotor performance (Elsworth *et al.* 2003). Similar seasonal reductions in dive duration have been observed in free-ranging turtles during summer (Carr *et al.* 1980; Bentivegna *et al.* 2003; Gordos *et al.* 2003*a*; Hochscheid *et al.* 2005; Bradshaw *et al.* 2007), and have also been shown to occur under experimental long-term thermal exposure (Clark *et al.* 2008), suggesting that inefficient thermal acclimatization of the physiological machinery responsible for diving may be common among ectothermic air-breathing divers. Further research is required to elucidate what feature of their diving physiology *C. johnstoni* were unable to thermally acclimatize, and to determine what constraints thermal-induced restrictions upon dive performance may place upon ecological function.

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