The Journal of Experimental Biology 206, 1171-1181

doi:10.1242/jeb.00230

Variability in brain and arterial blood temperatures in free-ranging ostriches in their natural habitat

Andrea Fuller[1,*], Peter R. Kamerman[1], Shane K. Maloney[1,2], Graham Mitchell[1] and Duncan Mitchell[1]

[1]*School of Physiology, University of the Witwatersrand Medical School, 7 York Road, Parktown 2193, South Africa* and [2]*Department of Physiology, School of Biomedical and Chemical Science, University of Western Australia, Crawley 6009, Perth, Australia*

*Author for correspondence (e-mail: fullera@physiology.wits.ac.za)

Accepted 10 January 2003

Summary

We used implanted miniature data loggers to measure brain (in or near the hypothalamus) and carotid arterial blood temperatures at 5 min intervals in six free-ranging ostriches *Struthio camelus* in their natural habitat, for a period of up to 14 days. Carotid blood temperature exhibited a large amplitude (3.0–4.6°C) circadian rhythm, and was positively correlated with air temperature. During the day, brain temperature exceeded carotid blood temperature by approx. 0.4°C, but there were episodes when brain temperature was lowered below blood temperature. Selective brain cooling, however, was not present in all ostriches, and was not tightly coupled to the prevailing body temperature. Brain temperature was maintained within narrow daily limits of approx. 2°C, and varied significantly less than blood temperature at short time scales of 5 to 20 min. At night, brain temperature exceeded blood temperature by as much as 3°C. We attribute the elevated brain temperatures to warming of cerebral arterial blood, by reduced heat exchange in the ophthalmic rete or possibly heat gain from cranial structures, before supplying the hypothalamus. Further studies are necessary to elucidate the significance of such variations in brain temperature and the importance of selective brain cooling in free-living birds.

Key words: thermoregulation, body temperature, selective brain cooling, circadian rhythm, bird, ostrich, *Struthio camelus*.

Introduction

The main determinant of deep brain temperature, measured in or near the hypothalamus, is the temperature of the arterial blood supplying the brain (Hayward and Baker, 1968). In most mammals, therefore, brain temperature changes in parallel with carotid blood temperature but, because of its high metabolic rate, exceeds the temperature of arterial blood by 0.2–0.5°C (Hayward and Baker, 1969). However, some mammals, particularly artiodactyls and felids, are able to lower brain temperature below carotid blood temperature, a process termed selective brain cooling (for reviews, see Jessen, 2001; Mitchell et al., 2002). In these mammals, selective brain cooling is facilitated by the carotid rete, a bilateral network of intertwining arteries in the main arterial supply to the brain, situated on either side of the pituitary body within cool venous lakes derived from veins draining the nasal mucosa (Gillilan, 1974; Simoens et al., 1987). Moderate exercise on a treadmill or exposure to heat in a laboratory uncouples brain and carotid blood temperatures, so that the gradient between brain and blood temperatures narrows until, at a threshold temperature of approximately 39°C, brain temperature equals carotid blood temperature (Baker, 1982; Kuhnen and Jessen, 1991; Kuhnen and Mercer, 1993). Beyond that threshold, brain temperature rises at a slower rate than does blood temperature, establishing selective brain cooling with a magnitude, at most, of about 1°C.

In mammals that are free-living in their natural habitat and exposed to a variety of complex stressors, however, a similar tight thermal relationship between brain and arterial blood temperatures is apparently absent. In free-ranging antelope, selective brain cooling occurs within the normothermic range of body temperature, and only sporadically in response to high heat loads (Jessen et al., 1994; Mitchell et al., 1997; Fuller et al., 1999b; Maloney et al., 2002). During high-intensity exercise, when brain temperatures reach their highest levels, selective brain cooling is abolished. Indeed, for any given blood temperature, brain temperature is highly variable and unpredictable. Large-amplitude, transient deviations in brain temperature, independent of any changes in the temperature of arterial blood supplying the brain, have also been observed in several laboratory animals in response to non-thermal stimuli (Fuller et al., 1999a; Maloney et al., 2001). This variability arises because the efferent arm of the control mechanism

underlying selective brain cooling is influenced by alterations in sympathetic nervous system activity (for a review, see Mitchell et al., 1987).

It is widely held that birds employ selective brain cooling, analogous to that in mammals. Most studies have shown that brain temperature in birds consistently is lower than core body temperature over a wide range of ambient and body temperatures (for a review, see Arad, 1990; Jessen, 2001). The anatomical structure thought to be responsible for this brain cooling is the ophthalmic rete, a network of extracranial arteries developed from the external ophthalmic branch of the internal carotid artery and closely associated with veins carrying cool blood away from the buccopharyngeal surfaces, beak and eyes (Richards, 1967; Kilgore et al., 1973). In contrast to mammals (Mitchell et al., 1987), however, no mechanism to control brain cooling in birds has been advanced. Moreover, the relationship between brain and carotid arterial blood temperature has not been described. Brain and carotid blood temperatures have been measured simultaneously in one study, but only sporadic measurements were obtained (Kilgore et al., 1973). As far as we are aware, continuous recordings of arterial blood temperature have not been made in any bird, and the variability of brain temperature over 24 h or longer has been measured in only a few species (Scott and van Tienhoven, 1971; Aschoff et al., 1973; Withers and Crowe, 1980). Thus, evidence for brain cooling in birds is derived from intermittent measurements of abdominal (colonic or cloacal) temperature. In mammals, however, abdominal (or rectal) temperatures usually overestimate arterial blood temperature (Bligh, 1957a). Their use as surrogates for arterial blood temperature may generate artefactual evidence for selective brain cooling in mammals (Maloney et al., 2001) and, it seems possible, also in birds. Another potential source of error arises from difficulties in measuring hypothalamic temperature. Temperatures obtained from sensors in short, large-diameter guide tubes may be contaminated by local ambient temperature (Fuller et al., 1998), raising doubts about the validity of some brain temperatures obtained from small birds. Brain temperatures also have never been continuously measured in any free-ranging bird in its natural habitat. Our recent studies of free-ranging mammals have shown that the thermoregulatory responses of animals in their natural environment cannot be predicted from measurements made on tame or restrained animals under laboratory conditions.

The aims of our study were to investigate the relationship between brain and arterial blood temperature in free-living birds in their natural habitat, and the variability in these temperatures in response to variations in thermal load. We hypothesized that brain temperature would be more variable than arterial blood temperature, and that selective brain cooling would be absent or of small magnitude. We report here our findings from six ostriches *Struthio camelus*, in which we implanted miniature data loggers with temperature probes to measure brain and carotid blood temperatures every 5 min.

Materials and methods

Animals and habitat

Experiments took place at the Lichtenburg Game Breeding Centre in Lichtenburg (26°07′S, 26°10′E), 220 km west of Johannesburg, in South Africa. The Centre extends over an area of 4500 ha, consisting mainly of grassland with scattered trees and shrubs. Experimental animals (see Table 1) consisted of three male and three female ostriches *Struthio camelus* L., which had been raised from hatching in an enclosure on the reserve, and were between 1 and 2 years old. The first group of animals (ostriches 1–3) was instrumented during December 1999, and an additional three animals (ostriches 4–6) were instrumented in January 2001 (both periods in southern hemisphere summer).

Surgery

All experimental procedures were approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (protocol no. 99/64/5). Ostriches were captured, blindfolded and transported to a nearby temporary surgical theatre. The animals were placed in sternal recumbency, supported by sandbags and inflated tyre tubes, and anaesthetised with 1–3% halothane (Fluothane, Zeneca) in oxygen, administered *via* an endotracheal tube. Respiratory rate, heart rate, blood pressure and colonic temperature were monitored throughout surgery.

Table 1. *Characteristics of each ostrich, and mean and amplitude of carotid and brain temperatures recorded every 5 min*

				Mean temperature over a 24 h period (°C)		Amplitude (peak-to-trough) (°C)	
Ostrich	*N*	Gender	Mass (kg)	Carotid	Brain	Carotid	Brain
1	8	Female	45	38.51±0.30	39.32±0.16	4.24±1.42	2.18±0.13
2	5	Female	40	38.45±0.20	39.21±0.07	3.17±1.10	2.00±0.37
3	6	Male	50	38.27±0.37	39.20±0.23	4.61±0.51	2.62±0.30
4	14	Male	55	38.52±0.66	38.68±0.38	3.06±1.02	2.02±0.32
5	5	Male	55	38.62±0.27	39.09±0.09	3.17±0.41	2.00±0.19
6	8	Female	45	38.18±0.25	38.94±0.30	4.62±0.53	1.98±0.10

Values are means ± S.D.; *N* = number of days of data.

Using aseptic surgical procedures, we implanted miniature data loggers and thermistors for temperature measurement. A thermistor in a blind-ended and thin-walled polytetrafluroethylene (PTFE) tube (0.9 mm o.d.; Straight Aortic Flush 4F Catheter, Cordis, The Netherlands) was inserted into the left common carotid artery at a position midway along the length of the neck, and advanced 100 mm into the artery, towards the heart. It was secured by a purse-string suture in the artery wall. Outside the artery, the PTFE tube was connected to rubber cable (approx. 150 mm length, 3 mm o.d.) containing leads from the thermistor to a temperature logger (see below). The logger, covered with an inert wax (Sasol, South Africa), was placed in a subcutaneous pouch near the artery. A second temperature logger, connected to a brain thermistor probe, was also positioned subcutaneously in the neck. Its cable was advanced subcutaneously to the skull, where it was connected to a head plate and guide tube. The guide tube, constructed from cellulose acetate butyrate tubing (30 mm length, 3.2 mm o.d., 1.6 mm i.d.; World Precision Instruments, Sarasota, USA) sealed at the tip by a steel cap, was positioned on the occipital skull, 5 mm distal to the parieto-occipital suture. Coordinates were determined from sections of dead ostriches of similar size, so that the probe tip would be positioned near the hypothalamus. The brain guide tube was connected to a small plastic headplate (10 mm×10 mm×3 mm), which was secured to the skull by two bone screws. In two ostriches (ostriches 5 and 6), we also measured abdominal temperatures, by inserting a thermistor 200 mm into the right side of coelomic cavity immediately behind the last rib. The thermistor (27-10K4A801, Onset Computer Corporation, Pocasset, USA), in silicone tubing, was connected to a logger that was positioned subcutaneously on the thorax. All equipment was positioned subcutaneously, with no external components.

A 50 mg enrofloxacin tablet (Baytril, Bayer) was placed in each surgical site and suture lines were sprayed with a topical antiseptic spray (Necrospray, Centaur Labs, Johannesburg). Each ostrich also received long-acting penicillin (10 ml i.m., Duplocillin, Intervet, Johannesburg), an opiate analgesic (buprenorphine, 0.3 mg i.m., Temgesic, Schering-Plough, Johannesburg) and an analgesic and anti-inflammatory medication (15 ml s.c., Dexa-Tomanol, Centaur Labs, Johannesburg). After surgery, animals were transported to a paddock. Recovery from anaesthesia was rapid (15–20 min). Thereafter the ostriches were released into a fenced 62 ha enclosure, where they ranged freely with several species of African mammals.

2 months after release, ostriches were herded into a paddock and individually captured. Using the same immobilisation, anaesthetic and surgical procedures as before, data loggers, headplates and thermistors were removed. All loggers positioned subcutaneously were in perfect order, wounds had healed, and there were no signs of infection. Examination of the carotid artery revealed no occlusion or clotting along the length of the intravascular guide tube, that is, thermistors measured the temperature of free flowing blood. In all ostriches, however, mechanical failure of at least one thermistor had occurred, usually as a result of breakage between 6 and 15 days after surgery. After recovery the ostriches were released back into the study enclosure and remain healthy.

Body temperature measurement

The miniature data loggers (StowAway XTI, Onset Computer Corporation, Pocasset, USA) had outside dimensions of approx. 50 mm×45 mm×20 mm and a mass of approx. 40 g, when covered in wax. These loggers were custom-modified for us, to have a storage capacity of 32 K, a measurement range from +34 to +46°C, and resolution of 0.04°C. The scan interval of the loggers was set at 5 min. Brain and blood temperature sensors were constructed from ruggedized glass-coated bead thermistors with insulated extension leads (bead diameter 0.3 mm; ABOE3-BR11KA103N, Thermometrics, Edison, USA). All temperature sensors were calibrated against a high-accuracy quartz thermometer (Quat 100, Heraeus, Hanau, Germany) in an insulated water bath, and had an accuracy of one sampling step of the logger (0.04°C).

Meteorological measurement

Climatic data were obtained from a portable weather station (Mike Cotton Systems, Cape Town, South Africa) at the study site. Rainfall was 86 mm during December 1999 and 106 mm during January 2001; total annual rainfall for the region was approx. 700 mm. Data on air temperature and wind speed are given in more detail below. Solar noon was at 13:00 h.

Data analyses

Animals were slightly hypothermic immediately after anaesthesia but warmed rapidly after release. Body temperature patterns on the day after surgery did not differ from those recorded for the remainder of the data collection period. To avoid introducing a circadian bias to body temperature analyses, we analysed data from midnight on the day following surgery to midnight before equipment failure for each animal. Analysed data consisted of between 5 and 14 days for the different ostriches (see Table 1).

To analyse variability in body temperatures over different time scales we performed a nested (hierarchical) analysis of variance (ANOVA) on carotid blood temperature and brain temperature. For this analysis, we used the first 5 days of data from each animal, to avoid introducing a bias associated with different environmental conditions in animals with larger data sets. The analysis was performed over the following time scales: over the full 5 days, over each day, over each hour, at 20 min intervals and at 5 min intervals. Total variability was equal to the variability of the 5 min readings. The analysis calculates the amount of variability introduced at each step up the hierarchy (for example, if the average daily carotid temperature for the ostriches was the same each day, no extra variance would be introduced at the level of ‘days within animals’). Variance for each subsequent level of the hierarchy includes the variance present in the lower level, so ‘added

variance' is calculated by subtraction for each level of the analysis. We determined statistical significance of added variance at each step using the variance ratio test. We also compared carotid temperature and brain temperature at each level of the hierarchy to determine whether the two temperatures varied in parallel.

Original 5 min recordings of body temperatures were used to find the daily mean, standard deviation (S.D.), minimum, maximum and amplitude of carotid blood temperature and brain temperature for each animal. The relationship between brain temperature and blood temperature in each animal was analysed by sorting all 5 min measurements of arterial blood temperature into 0.1°C classes, and determining the mean, standard deviation, maxima and minima of brain temperature at each class of blood temperature. The frequencies at which each of the 0.1°C classes of blood temperature occurred were also determined.

Hourly means of blood and brain temperatures were compared to 1 h measurements of meteorological variables using linear correlation (Pearson product-moment) and regression analysis. $P<0.05$ was set as the minimum acceptable level of statistical significance.

Results

Fig. 1 shows mean 5 min brain and carotid blood temperatures, as a function of time of day, for one ostrich over a period of 8 days. In this animal, and in all other birds, mean daily brain temperature exceeded mean carotid blood temperature (see also Table 1). However, the difference between brain and arterial blood temperature was not constant. During the day, when both temperature rhythms exhibited a plateau-like peak (from 09:00 h to 18:00 h), brain temperature, on average in all birds, exceeded blood temperature by 0.4°C. At night this difference increased, with mean carotid blood temperature dropping by up to 2°C more than mean brain temperature. As a result, the amplitude of the blood temperature rhythm, calculated as the difference between the maximum and minimum recorded temperature on each day, was significantly higher than that of brain temperature (Student's paired *t* test, $P<0.001$ for all animals). Indeed, blood

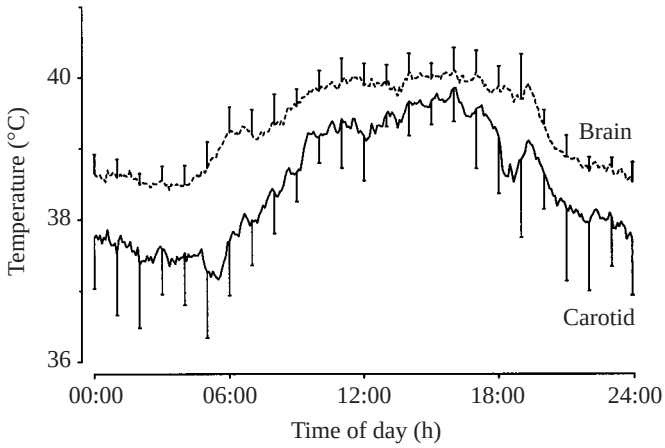

Fig. 1. Brain and carotid blood temperatures, as a function of time of day, for ostrich 1 over a period of 8 days. Values are means ± S.D. (shown at 1 h intervals) of 5 min readings.

temperature, on average, fluctuated over a daily range twice as great as that of brain temperature (Table 1).

We further analysed differences in variability between brain and arterial blood temperature, at different time scales, by carrying out a nested (hierarchical) ANOVA, the results of which are shown in Table 2. For both carotid temperature and brain temperature, significant variability was added at the first two levels of our hierarchy ('20 min within hours' and 'hours within days'). The greatest addition in variability was introduced at the level 'hours within days', presumably as a result of the marked 24 h oscillation in body temperature evident in Fig. 1. No extra variability in carotid temperature was added at the levels of 'days within animals' and 'between animals', indicating that mean daily carotid temperature for each animal did not differ over the data collection period, and that each animal experienced similar mean 24 h carotid temperatures. Although brain temperatures did not differ within an animal across days, there were differences in mean brain temperatures between animals. These differences may reflect episodes of brain cooling in some birds but not in others (see below for further details).

Comparison of carotid temperature and brain temperature at

Table 2. *Results of the nested (hierarchical) analysis of variance on brain and carotid blood temperature of six ostriches*

	[a]Variance ratio				[b]Carotid *versus* brain	
Source of variation	Carotid		Brain		Higher variability added	
Between animals	0.7	NS	5.0	***	Brain	NS
Between days within animals	0.1	NS	0.1	NS	Carotid	**
Between hours within days	22.6	***	22.7	***	Carotid	***
Between 20 min blocks within hours	1.9	***	1.8	***	Carotid	***
Between 5 min recordings within 20 min	1		1		Carotid	***

[a]Variance ratio values show the significance of each level of the hierarchy for the two body temperatures.

[b]The site of temperature measurement to which most variability was added at each level and the significance of the difference between the amount of variability added to each temperature.

NS, not significant; **$P<0.01$; ***$P<0.001$.

each level of the hierarchy, using the overall total sum of squares, revealed that there was significantly more variation in carotid temperature than in brain temperature, at the four lowest levels of the hierarchy. Brain temperature, therefore, was more stable from day to day, from hour to hour, and from minute to minute, than was carotid temperature. At the highest level of 'between animals', there was no significant difference in variation added between carotid temperature and brain temperature, indicating that variability between overall means of the animals was similar for the two temperatures. In summary, these analyses show that mean carotid temperature was similar, from day to day, within individual animals, and between animals, but was significantly more variable overall than was brain temperature. Brain temperature varied significantly less than carotid temperature at short time scales (minutes and hours), but patterns of brain temperature differed between animals.

The data in Fig. 1 and Table 1 are averages, so they mask short-term oscillations in body temperature, and conceal significant relationships between blood and brain temperatures. Fig. 2 shows the original records of body temperatures from two animals at 5 min intervals, during periods when large, short-duration decrements in blood temperature occurred simultaneously in all animals in each flock. We believe that these falls, which ranged from 2 to 4°C in magnitude and lasted for up to 2 h, represent episodes of drinking. There were no unusual climatic conditions during these times, and similar falls in carotid temperature were seen on a few other non-consecutive days, also in the evening. Regardless of the cause, brain temperature did not parallel changes in blood temperature, but dropped by a much smaller amount, or even remained constant.

Other smaller magnitude decrements in carotid blood temperature, which occurred frequently and presumably as a consequence of events other than drinking, were also not associated with changes in brain temperature. Typical oscillations in body temperatures, on one day, are shown for one ostrich (ostrich 1) in Fig. 3. On several occasions, carotid temperature dropped by up to 1°C in less than 1 h, but brain temperature remained constant or decreased by only a few tenths of a degree Celsius. In particular, brain temperature remained remarkably constant in all animals during the night, when large short-duration oscillations in carotid temperature were often evident.

Although brain temperature was higher than carotid blood temperature for most of the day, and always at night, Fig. 3 also shows an episode in the afternoon when brain temperature was lower than blood temperature. Similar episodes of selective brain cooling were observed routinely in two ostriches, but only occasionally in the other four animals. In most instances, selective brain cooling resulted from rises in carotid temperature that were not accompanied by similar rises in brain temperature. The selective brain cooling that did occur tended to happen when the ostriches had a high body temperature; however, high body temperatures were not always accompanied by selective brain cooling.

Fig. 4 shows brain temperature as a function of arterial blood temperature, and the frequency distribution of blood temperature in four ostriches. Data from ostriches 2 and 6, omitted for clarity, were similar to those obtained for ostrich 1 (Fig. 4Ai). The diagonal lines in the upper panels show the line of identity of brain and blood temperature. In all animals, brain temperature exceeded carotid blood temperature, sometimes by more than 3°C, at carotid temperatures less than 38°C. The slopes of lines fitted to data points at carotid temperatures of less than 38°C were not significantly different to zero, indicating that brain temperature was independent of

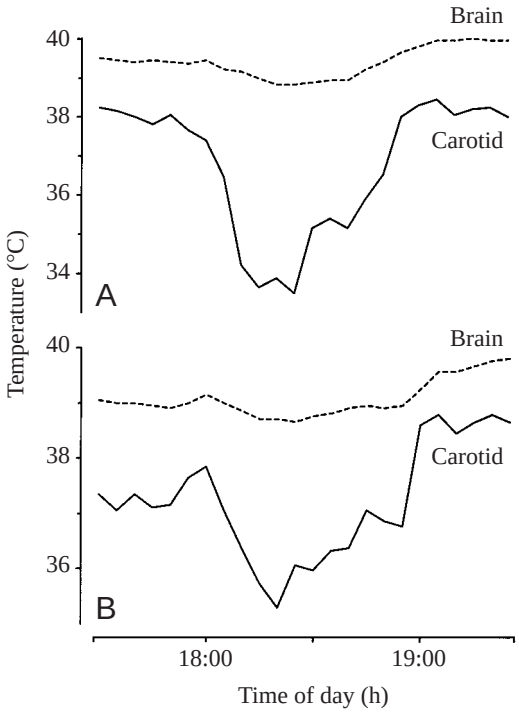

Fig. 2. Original records of brain and carotid blood temperatures taken at 5 min intervals from two ostriches (ostrich 1, A; ostrich 3, B) over a 2 h period on the same day.

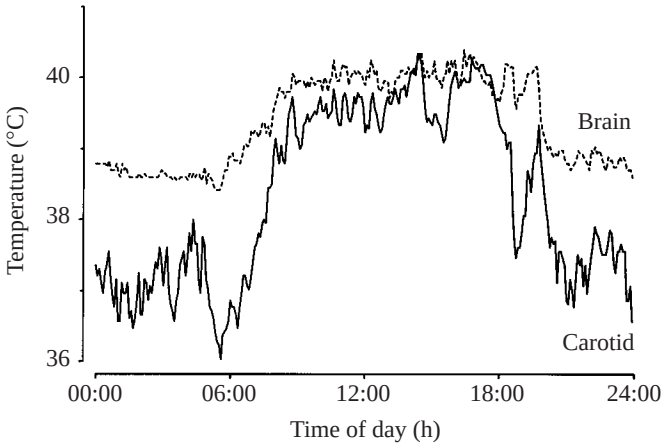

Fig. 3. Original records of brain and carotid blood temperatures taken at 5 min intervals from ostrich 1 over 1 day.

carotid temperature at these low body temperatures. At higher body temperatures, mean brain temperature at each carotid temperature remained above the line of identity, except in ostrich 4 (Fig. 4Di). This animal was unusual, in that mean brain temperature was lower than mean carotid temperature at carotid temperatures between 38.4 and 39.2°C. However, inspection of the frequency distribution of carotid blood temperature shows that the animal infrequently experienced

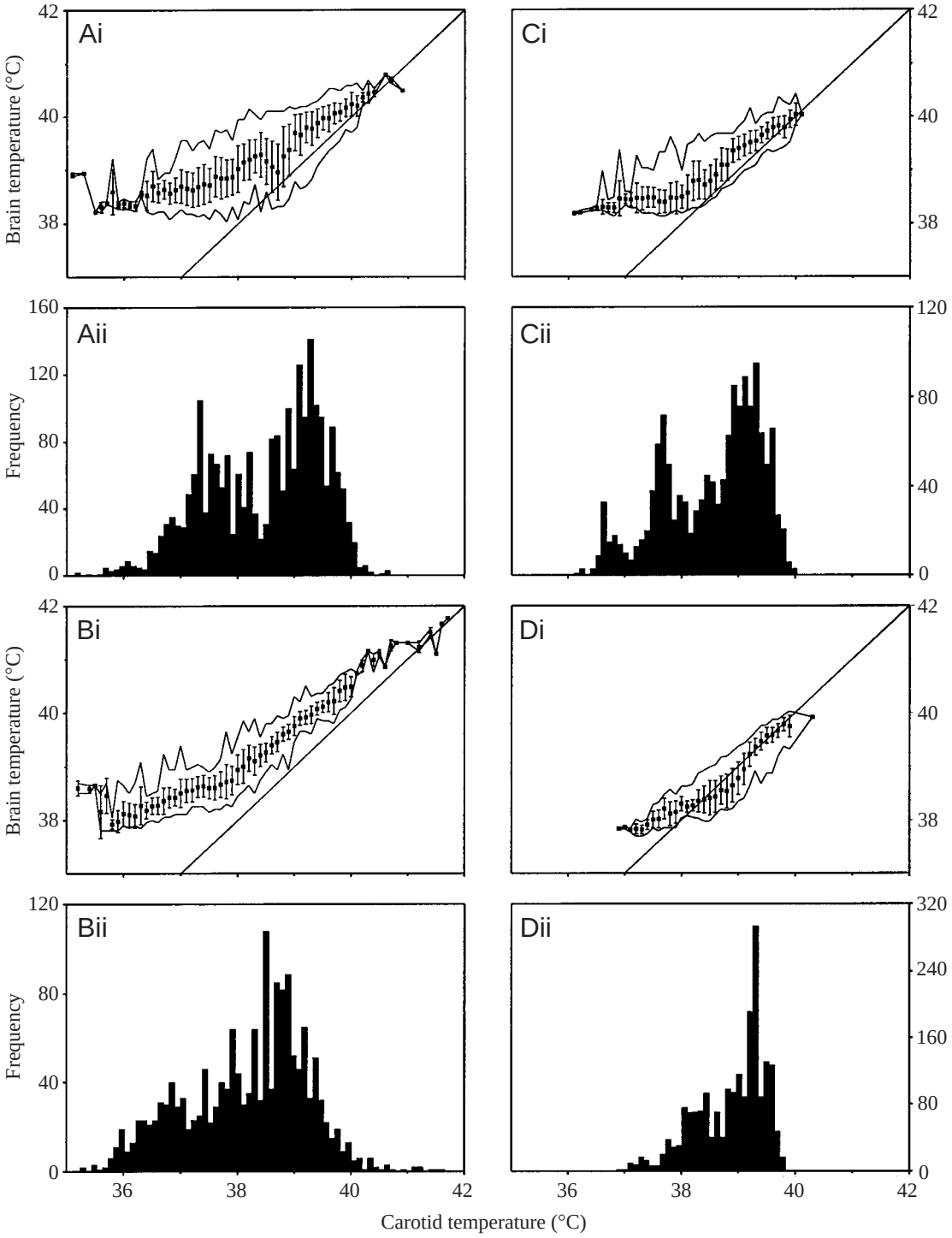

Fig. 4. Brain temperature as a function of carotid blood temperature (Ai–Di), and frequency distribution of blood temperature (Aii–Dii), in four ostriches. The 5 min values of blood temperature were sorted into classes of 0.1°C width. Values in Ai–Di are means ± S.D. and minima and maxima of brain temperature at each class of blood temperature. Aii–Dii show absolute frequencies at which each class of blood temperature occurred; *N*=2304 data points for ostrich 1 (A), 1728 for ostrich 3 (B), 1440 for ostrich 5 (C) and 4032 for ostrich 4 (D). The lines of identity of brain and blood temperatures are shown in Ai–Di.

carotid temperatures in this range. At its mode of body temperature (39.3°C), mean brain temperature was slightly higher than blood temperature. The same pattern was true for all six ostriches, despite differences in the frequency distribution of blood temperatures. Results obtained from ostrich 3 (Fig. 4Bi) were also atypical, in that this animal never exhibited selective brain cooling (except for one isolated 5 min measurement of body temperature).

Differences in body temperature patterns of animals may reflect differences in size, age or gender of the animals (see Table 1), and also differences in climatic conditions. Maximum air temperatures and solar radiation levels in the first study (ostriches 1–3) were significantly lower than those recorded a year later (ostriches 4–6). In December 1999, mean air temperature fluctuated between 13°C and 29°C; in January 2001 it ranged from 11°C to 37°C. Wind speeds in both periods were similar, fluctuating daily between 2 and 4 m s^{-1}, on average, and reaching highest levels at approx. 08:00 h. Part of the variability in internal body temperatures may be attributed to variations in the environmental thermal load. In each of the six animals, brain and carotid blood temperatures, averaged over 1 h periods, were significantly correlated with air temperature (Pearson product-moment correlation; r=0.56–0.83 for brain temperatures, r=0.64–0.85 for carotid temperatures, P<0.0001 in all cases). Slopes of linear regression lines fitted to air temperature and carotid temperature for each animal ranged from 0.08°C to 0.21°C, so that, on average, carotid blood temperature of ostriches increased 0.15°C per 1°C increase of air temperature. The slopes of similar regression lines between air temperature and brain temperature were lower (0.06–0.14°C, mean 0.10°C), reflecting the relatively stability of brain temperature in response to variations in carotid temperature.

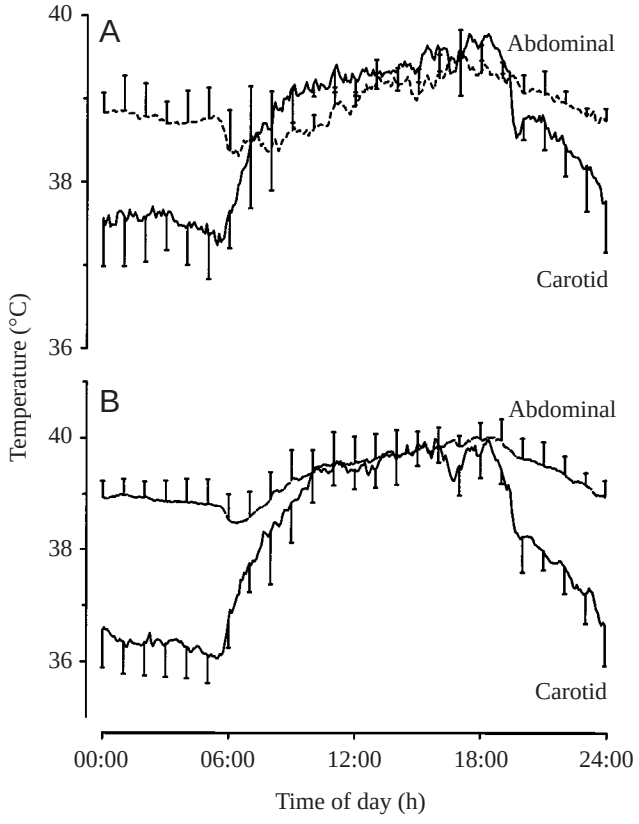

Fig. 5. Abdominal and carotid blood temperatures, as a function of time of day, for two ostriches over 5 days (ostrich 5, A) or 8 days (ostrich 6, B). Values are means ± S.D. (shown at 1 h intervals) of 5 min readings.

Previous studies of thermoregulation in birds have measured cloacal or abdominal temperature as an index of core body temperature, rather than arterial blood temperature. We therefore compared carotid blood temperature with abdominal temperature, in two birds. In both animals, the nychthemeral amplitude of abdominal temperature (1.75±0.57°C, ostrich 5; 1.79±0.29°C, ostrich 6; means ± S.D.) was significantly less than that of carotid temperature (Student's paired t-test, P<0.001; see Table 1). Abdominal temperature always exceeded carotid blood temperature at night, often by as much as 3°C (Fig. 5). However, the relationship between abdominal temperature and carotid blood temperature during the day was not clear. In one animal (ostrich 5), mean carotid blood temperature consistently exceeded mean abdominal temperature by approx. 0.4°C (Fig. 5A). In the second ostrich, mean daytime abdominal temperature was equal to, or slightly higher than, mean carotid temperature (Fig. 5B). These differences probably reflect different thermal gradients at different sites in the ostrich coelomic cavity.

Discussion

We have obtained the first continuous measurements of brain and arterial blood temperatures in any bird, free-living in its natural habitat. Our results show that ostriches maintained brain temperatures within a narrow daily range of approximately 2°C, and that brain temperature fluctuated less over short time scales (hours or minutes) than did the temperature of the arterial blood supplying the head. Patterns of brain temperature, however, differed between animals, with some ostriches occasionally exhibiting brain temperatures lower than carotid blood temperature, but others rarely employing such selective brain cooling. Although selective brain cooling tended to be present at high body temperatures, during the afternoon, there was no clear relationship between body temperature and the implementation of selective brain cooling. In contrast to brain temperature, carotid blood temperature was relatively constant across days within individual animals, and between animals. However, carotid temperatures were strongly correlated with environmental thermal load, and fluctuated daily by as much as 4.6°C.

The amplitude of this nychthemeral oscillation in carotid blood temperature of ostriches is more than twice that measured in free-ranging antelope and zebras occupying a similar habitat (Jessen et al., 1994; Mitchell et al., 1997; Fuller et al., 1999b, 2000; Maloney et al., 2002). It is also significantly greater than the range of abdominal temperature

measured in two ostriches in this study (Fig. 5), and higher than colonic temperature fluctuations reported previously for ostriches (Bligh and Hartley, 1965; Louw et al., 1969) and other birds (Refinetti and Menaker, 1992). These differences raise concerns about the reliability of arterial blood temperature measurements in our study. Indeed, most investigators use surrogates of blood temperature because it is technically difficult to measure blood temperature accurately, particularly in small animals. Previous investigators, for example, have reported occlusion of the carotid artery along the intravascular part of the guide tube (Jessen et al., 1998). However, we are confident that we measured the temperature of free-flowing blood in the carotid artery. On removal of instruments, we observed the tips of thermistor probes floating freely in the blood stream, and no impedance to blood flow. We also have used the same technique to measure blood temperatures accurately in other species with smaller diameter vessels (for example pigs; Fuller et al., 1999a). Moreover, if thermistors were enclosed in scar tissue rather than in flowing blood, our measurements would probably have underestimated true variability in blood temperature.

Another possibility is that carotid blood temperature differs from the temperature of arterial blood leaving the heart. However, since blood flow is high and the carotid artery is positioned deep in the neck where it is well insulated, arterial temperatures measured near the heart are likely to be identical to those at the base of the brain (Hayward and Baker, 1968). Indeed, Kilgore et al. (1973) showed that arterial blood temperatures measured near the brain and near the heart did not differ in rheas, despite the birds having long necks. In some mammals, however, a difference in the temperature between the right and left sides of the heart may exist. Usually, inspired air is heated and humidified by the upper airways, so that it is fully saturated with water vapour and at body temperature when it reaches the alveoli (McFadden, 1983). However, if ventilation rate is sufficiently high, unsaturated air reaches the alveoli and the lungs become a potential site of evaporative heat loss. In exercising horses, blood temperature decreased by as much as 0.6°C on its passage through the lungs, dissipating up to two-thirds of the total heat load generated (Hodgson et al., 1993; Lund et al., 1996). The temperature of blood in the left atrium or carotid trunk also has been found to be lower than pulmonary artery temperature in resting dogs (Mather et al., 1953) and calves (Bligh, 1957b), albeit by a much smaller amount than that in exercising horses.

Heat loss from respiratory surfaces may explain some of the variability in carotid temperatures of ostriches. Pulmonary capillary blood in the ostrich lung is exposed to air over a large respiratory surface area (Maina and Nathaniel, 2001). An increase in ventilation rate of the lung, in response to a heat load or other non-thermal stressors, might increase the gradient between pulmonary artery and carotid blood temperature, leading to a fall in carotid temperature. Such heat loss might explain why carotid blood temperature, contrary to expectations for a large mass bird, frequently demonstrated small-amplitude (approx. 1°C) oscillations over short time periods (Fig. 3). Similar deviations were not seen in abdominal temperatures, presumably because of high thermal inertia in the body cavity or localised warming. The largest decrements in carotid temperature that we observed, usually in the evening (Fig. 2), were probably associated with drinking. Similar rapid falls in carotid blood temperature have been recorded in goats after drinking (Jessen et al., 1998) and in free-ranging springbok (Mitchell et al., 1997). Prolonged falls at night probably represent whole body cooling, associated with low air temperatures. Carotid blood temperatures of ostriches, unlike those obtained from free-ranging antelope, were positively correlated with ambient temperature.

In the face of high short-term variability in carotid blood temperature, brain temperature remained remarkably constant. These findings are at odds with data obtained from large mammals. Analysis of variability in body temperatures of free-ranging oryx, using the same statistical techniques as those performed in this study, revealed that brain temperature varied significantly more, over time scales of 5–20 min, than did arterial blood temperature (Maloney et al., 2002). Indeed, short-term variability in brain temperature in response to a variety of non-thermal stimuli has been demonstrated in several mammals that possess a carotid rete, including pigs (Fuller et al., 1999a) and sheep (Maloney et al., 2001). These variations mainly reflect the dynamic influence of sympathetic nervous system output on the supply of cool venous blood to the venous lakes surrounding the carotid rete (Bamford and Eccles, 1983). Although short-term dissociations in brain and arterial blood temperature of mammals may occur in this way, however, hypothalamic temperature usually changes in the same direction as carotid blood temperature, and rarely differs from it by more than 0.5°C. In ostriches, however, brain temperature frequently varied independently of carotid temperature, particularly when carotid blood temperatures fell. During the day, when blood temperature was in the 38–40°C range, brain temperature exceeded blood temperature by approx. 0.4°C, as it does in mammals at most times of the nychthemeral rhythm. However, at night this difference increased to as much as 3°C (Fig. 3), a magnitude greater than that previously observed in any other animal. This remarkable temperature difference may reflect a unique anatomical basis underlying brain temperature regulation in ostriches.

In birds, there are two parallel routes *via* which arterial blood may reach the brain (Richards, 1970; Kilgore et al., 1973). The first is a direct route *via* the cerebral arteries that arise from an incomplete circle of Willis, or intercarotid anastomosis (Baumel and Gerchman, 1968), at the base of the brain. The anastomosis, together with sections of the internal carotid and internal ophthalmic arteries, lies within the cavernous sinus, which receives venous blood from the ethmoid and superficial ophthalmic veins draining the head. Blood that reaches the hypothalamus *via* this route should have a temperature similar to that of carotid arterial blood, although Richards (1970) has suggested that the close association between veins and arteries at the base of the brain may facilitate some heat exchange. The second route is indirect, *via* the ophthalmic rete, an

intermingled network of arteries arising from the external ophthalmic artery, which is an extracranial branch of the internal carotid artery. Arteries distal to the rete supply the eye, and also anastomose with intracranial branches of the internal carotid to supply the brain (Kilgore et al., 1976). Functionally, the ophthalmic rete appears to be an analogue of the carotid rete in mammals, allowing heat to be transferred from warm arterial blood to cool venous blood returning from evaporative surfaces of the head. Blood from the two pathways is thought to mix, so that the resultant brain temperature reflects the relative contribution of blood from the each pathway (Midtgard, 1983). If arterial blood is cooled in the rete, and most of the blood supply to the brain is *via* this indirect pathway, selective brain cooling will be invoked. There is convincing evidence that birds employ the ophthalmic rete in this manner (for a review, see Arad, 1990). Blocking blood flow to the rete (Kilgore et al., 1979) or impairing evaporative cooling of cranial venous blood (Bernstein et al., 1979) reduces, or even reverses, the positive body-to-brain temperature difference. Moreover, birds that have a poorly developed rete (zebra finch; Bech and Midtgard, 1981) or no rete (calliope hummingbird; Burgoon et al., 1987), exhibit a reduced capacity for selective brain cooling. Although the cranial blood supply in ostriches has not been systematically investigated, it has been reported that they have a well-developed ophthalmic rete (Midtgard, 1983), and one would predict that selective brain cooling occurs by a similar mechanism.

In contrast to reports for other bird species, however, free-ranging ostriches exhibited selective brain cooling only sporadically. It may be that most of the blood supply to the brain bypasses the ophthalmic rete. However, we believe that it is also possible that birds do not implement selective brain cooling routinely, and that the reported brain cooling in birds is an experimental artefact. As a consequence of difficulties associated with measuring arterial blood temperature, most studies tendering support for brain cooling in birds have used cloacal or abdominal temperature as a surrogate. In mammals, however, the relationship between abdominal temperature and carotid blood temperature is not predictable, and comparison of rectal temperature and brain temperature leads to the erroneous conclusion that sheep routinely use selective brain cooling in cool and thermoneutral environments (Maloney et al., 2001). Indeed, we showed that abdominal temperature of ostriches was similar to carotid blood temperature during the day, but at night temperatures at the two sites differed by up to 3°C (Fig. 5). Abdominal temperatures in a large bird like the ostrich are likely to respond slowly to changes in thermal status of the body, and also are unlikely to be uniform at different sites in the large coelomic cavity. We measured abdominal temperatures in only two birds and thus do not want to speculate too much on the significance of the differences in carotid blood and abdominal temperature in ostriches. Kilgore et al. (1973) reported that carotid blood temperature of rheas was almost identical to cloacal temperature, but they obtained measurements only at ambient temperatures above 30°C, from three tame, restrained birds. Other reports of selective brain cooling in birds also may have resulted from comparison between brain temperature and a stable, warm abdominal temperature, rather than a variable and cooler carotid artery temperature.

The view that birds constantly employ selective brain cooling, with a magnitude of about 1°C, also is difficult to reconcile with the anatomical basis underlying selective brain cooling. Not only does the direct route of the internal carotid artery to the brain provide a pathway for arterial blood to bypass the rete, but there also is a shunt *via* which both arterial and venous blood may bypass the rete (Midtgard, 1983). Channelling blood *via* these different routes conceivably offers a way to regulate brain temperature. However, although brain temperature in birds appears to be remarkably constant, no evidence for any control mechanism has been advanced. We cannot explain why some of our ostriches employed selective brain cooling more frequently than did others. Although selective brain cooling occurred more often at high body temperatures, there was no clear threshold at which selective brain cooling was evoked. However, this variability between animals and the weak relationship between thermal status of the body and selective brain cooling is also evident in free-ranging antelope (Jessen et al., 1994; Mitchell et al., 1997; Fuller et al., 1999b; Maloney et al., 2002), and probably reflects the influence of multiple inputs on thermoregulatory effectors. The chief difference between free-ranging ostriches and antelope is the pattern of brain temperature regulation at night.

Indeed, in ostriches it appears unlikely that a significant proportion of arterial blood reaches the hypothalamus *via* the direct route of the internal carotid artery. If that were the case, we would expect brain temperature to closely track changes in carotid blood temperature at night. Richards and Sykes (1967) have demonstrated that the indirect route *via* the rete provides an adequate supply of blood to the brain if the direct route is occluded. If arterial blood is directed mainly *via* this indirect pathway to the brain, then there are three possible ways in which the large positive gradient between brain temperature and blood temperature may be established: (1) by increased metabolic heat generation in brain tissue; (2) by a decrease in cerebral blood flow; and (3) by warming of cerebral arterial blood supplying the hypothalamus. Of these three options, we believe that the first two are unlikely to account for the temperature difference. Brain heat production, at least in mammals, is tightly coupled to cerebral blood flow, so any increase in metabolic rate is matched by a similar increase in blood flow that removes additional heat and prevents a rise in brain temperature (Hayward and Baker, 1968). A decrease in cerebral blood flow, without a change in metabolic heat production, would reduce clearance of heat from the brain and lead to an increased brain temperature. However, even cessation of cerebral blood flow (Hayward and Baker, 1968) is unlikely to be sufficient to establish a 3°C rise in brain temperature. In mammals, brain temperature changes evoked by a wide variety of stimuli can all be explained by a prior shift

in the temperature of cerebral arterial blood (Hayward and Baker, 1968). We believe therefore that it is more likely that cerebral blood is warmed, as it transverses structures in the head, *en route* to the hypothalamus.

If blood supply to the hypothalamus is primarily *via* the indirect ophthalmic rete pathway, then an increase in the temperature of venous blood bathing the rete at night would increase the gradient between brain and blood temperature. Certainly, we would expect vasoconstriction of peripheral blood vessels and a marked reduction in evaporative heat loss from the head during the cold night, when ostriches are inactive (Williams et al., 1993). Heat loss from the head may be further reduced if the eyes of the bird are closed. Pinshow et al. (1982) showed that heat loss from the eye plays a significant role in reducing brain temperature of pigeons. The ostrich has a relatively large eye, which potentially is a large heat sink for arterial blood supplying the brain. Other possibilities, more likely to account for the large difference between brain and blood temperature, are that arterial blood destined for the hypothalamus exchanges heat with warm blood leaving the brain, or circulates through other brain tissues and is progressively warmed by the heat of local neural metabolism. However, it is not clear how flow *via* such routes could be regulated to achieve a reduced brain–blood temperature gradient during the day. Further studies of the anatomical basis underlying brain temperature regulation in ostriches are needed.

Similar brain warming at night has not been reported previously in birds or mammals, although it has been hypothesised that brain warming occurs during REM sleep in mammals (Wehr, 1992). Penguins have a well-developed ophthalmic rete that may serve the function of reducing heat loss from the poorly insulated head, in so doing keeping the brain and eyes warm (Frost et al., 1975). Similarly, there is evidence that at least 20 species of fishes and sharks use a heat-producing tissue or countercurrent heat exchanger to elevate brain and eye temperatures above that of the rest of the body, and the water temperature (Block, 1986). The function of such brain warming is unclear, but it may improve neural function in the face of rapid body cooling.

The role of selective brain cooling in birds has also not been resolved. For many years it was thought that selective brain cooling, in mammals or birds, functions to protect the apparently vulnerable brain from thermal damage during heat stress (for a review, see Mitchell et al., 1987). However, recent studies of free-ranging mammals in their natural habitat have yielded data that are incompatible with that concept (for reviews, see Jessen, 2001; Mitchell et al., 2002). Rather than being a process that favours protection of the brain, our current view is that selective brain cooling plays a role in whole body thermoregulation (Jessen, 2001; Mitchell et al., 2002). By cooling the hypothalamus, selective brain cooling reduces the drive on evaporative heat loss effectors, in so doing saving body water. If the role of selective brain cooling is indeed to balance thermoregulatory and osmoregulatory functions in this manner, then what is its role in birds? Unlike mammals, hypothalamic thermosensitivity plays a negligible role in adjusting autonomic output in birds, particularly in the hypothermic range of hypothalamic temperature (Jessen, 1996), so it is doubtful that selective brain cooling serves to adjust heat loss mechanisms. There also is no evidence that selective brain cooling in birds is controlled. Ostriches seldom employed selective brain cooling, and its implementation was unpredictable. Indeed, it may be that selective brain cooling in ostriches serves no current physiological function. Caputa et al. (1998) recently suggested that selective brain cooling plays a role in protecting the avian brain from asphyxic damage during diving, and that such brain cooling is an active and controlled mechanism. Further measurements of carotid blood and hypothalamic temperature in other species, particularly those of small body mass, are needed to accurately describe the relationship between brain and carotid arterial blood temperatures in birds. Moreover, additional investigations in free-ranging birds, including diving birds, using remote temperature-recording techniques are essential if we are to understand the significance of brain temperature patterns in birds.

We thank Dr Ferdi Schoeman and the National Zoological Gardens of South Africa for allowing us access to their property and resources, and the staff at the Lichtenburg Game Breeding Centre for their help. In particular, we thank the Head of the Game Breeding Centre, Andre Matthee, whose continued excellent support and animal capture skills made this project viable. We also thank Sr Mary-Ann Costello for assistance with surgical procedures, and Fiona Baker, Raymond Cherry, Alida Faurie, David Gray, Rachel Gray and Samantha Gray for their help on site. We also are indebted to Peter Karner at the Oryx Ostrich Abattoir for providing ostrich specimens, and Hank Burchinger at Onderstepoort for supplying an ostrich for a pilot study. This work was funded by the National Research Foundation, South Africa, a University Research Committee Grant, and a Medical Faculty Research Endowment Fund award, University of the Witwatersrand.

References

Arad, Z. (1990). Avian brain cooling – a review. *J. Basic. Clin. Physiol. Pharmacol*. **1**, 241-254.

Aschoff, C., Aschoff, J. and Saint Paul, U. v. (1973). Circadian rhythms of chicken brain temperatures. *J. Physiol*. **230**, 103-113.

Baker, M. A. (1982). Brain cooling in endotherms in heat and exercise. *Ann. Rev. Physiol*. **44**, 85-96.

Bamford, O. S. and Eccles, R. (1983). The role of sympathetic efferent activity in the regulation of brain temperature. *Pflügers Arch*. **396**, 138-143.

Baumel, J. J. and Gerchman, L. (1968). The avian intercarotid anastomosis and its homologue in other vertebrates. *Amer. J. Anat*. **122**, 1-18.

Bech, C. and Midtgard, U. (1981). Brain temperature and the rete mirabile ophthalmicum in the Zebra Finch (*Poephila guttata*). *J. Comp. Physiol*. **145**, 89-93.

Bernstein, M. H., Sandoval, I., Curtis, M. B. and Hudson, D. M. (1979). Brain temperatures in pigeons: effects of anterior respiratory bypass. *J. Comp. Physiol.* **129**, 115-118.

Bligh, J. (1957a). The relationship between the temperature in the rectum and of the blood in the bicarotid trunk of the calf during exposure to heat stress. *J. Physiol.* **136**, 393-403.

Bligh, J. (1957b). A comparison of the temperature of the blood in the pulmonary artery and in the bicarotid trunk of the calf during thermal polypnoea. *J. Physiol.* **136**, 404-412.
Bligh, J. and Hartley, T. C. (1965). The deep body temperature of an unrestrained ostrich *Struthio camelus* recorded continuously by a radio-telemetric technique. *Ibis* **107**, 104-105.
Block, B. A. (1986). Structure of the brain and eye heater tissue in marlins, sailfish, and spearfishes. *J. Morph.* **190**, 169-189.
Braasch, D. (1964). Zur Pathogenese des tödlichen Kreislaufkollapses nach Überwarmung einzelner Organe auf 45°C. *Pflügers Arch.* **278**, 567-574.
Brengelmann, G. L. (1993). Specialised brain cooling in humans? *FASEB J.* **7**, 1148-1153.
Burgoon, D. A., Kilgore, D. L. and Motta, P. J. (1987). Brain temperature in the calliope hummingbird (*Stellula calliope*): a species lacking a rete mirabile ophthalmicum. *J. Comp. Physiol. B* **157**, 583-588.
Caputa, M., Folkow, L. and Blix, A. S. (1998). Rapid brain cooling in diving ducks. *Am. J. Physiol.* **275**, R363-R371.
Frost, P. G. H., Siegfried, W. R. and Greenwood, P. J. (1975). Arterio-venous heat exchange systems in the Jackass penguin *Speniscus demersus. J. Zool. Lond.* **175**, 231-241.
Fuller, A., Carter, R. N. and Mitchell, D. (1998). Brain and abdominal temperatures at fatigue in rats exercising in the heat. *J. Appl. Physiol.* **84**, 877-883.
Fuller, A., Maloney, S. K., Kamerman, P. R., Mitchell, G. and Mitchell, D. (2000). Absence of selective brain cooling in free-ranging zebras in their natural habitat. *Exp. Physiol.* **85**, 209-217.
Fuller, A., Mitchell, G. and Mitchell, D. (1999a). Non-thermal signals govern selective brain cooling in pigs. *J. Comp. Physiol. B* **169**, 605-611.
Fuller, A., Moss, D. G., Skinner, J. D., Jessen, P. T., Mitchell, G. and Mitchell, D. (1999b). Brain, abdominal and arterial blood temperatures of free-ranging eland in their natural habitat. *Pflügers Arch.* **438**, 671-680.
Gillilan, L. A. (1974). Blood supply to brains of ungulates with and without a rete mirabile caroticum. *J. Comp. Neurol.* **153**, 275-290.
Hayward, J. N. and Baker, M. A. (1968). Role of cerebral arterial blood in the regulation of brain temperature in the monkey. *Am. J. Physiol.* **215**, 389-403.
Hayward, J. N. and Baker, M. A. (1969). A comparative study of the role of the cerebral arterial blood in the regulation of brain temperature in five mammals. *Brain Res.* **16**, 417-440.
Hodgson, D. R., McCutcheon, L. J., Byrd, S. K., Brown, W. S., Bayly, W. M., Brengelmann, G. L. and Gollnick, P. D. (1993). Dissipation of metabolic heat in the horse during exercise. *J. Appl. Physiol.* **74**, 1161-1170.
Jessen, C. (1996). Interaction of body temperatures in control of thermoregulatory effector mechanisms. In *Handbook of Physiology: Environmental Physiology*, vol. I (ed. M. J. Fregly and C. M. Blatteis), pp. 127-138. New York: Oxford University Press.
Jessen, C. (2001). Selective brain cooling in mammals and birds. *Jpn. J. Physiol.* **51**, 291-301.
Jessen, C., Dmi'el, R., Choshniak, I., Ezra, D. and Kuhnen, G. (1998). Effects of dehydration and rehydration on body temperatures in the black Bedouin goat. *Pflügers Arch.* **436**, 659-666.
Jessen, C., Laburn, H. P., Knight, M. H., Kuhnen, G., Goelst, K. and Mitchell, D. (1994). Blood and brain temperatures of free-ranging black wildebeest in their natural environment. *Am. J. Physiol.* **267**, R1528-R1536.
Kilgore, D. L., Bernstein, M. H. and Schmidt-Nielsen, K. (1973). Brain temperature in a large bird, the rhea. *Am. J. Physiol.* **225**, 739-742.
Kilgore, D. L., Bernstein, M. H. and Hudson, D. M. (1976). Brain temperatures in birds. *J. Comp. Physiol. B* **110**, 209-215.
Kilgore, D. L., Boggs, D. F. and Birchard, G. F. (1979). Role of the rete mirabile ophthalmicum in maintaining the body-to-brain difference in pigeons. *J. Comp. Physiol.* **129**, 119-122.
Kuhnen, G. and Jessen, C. (1991). Threshold and slope of selective brain cooling. *Pflügers Arch.* **418**, 176-183.
Kuhnen, G. and Mercer, J. B. (1993). Selective brain cooling in resting and exercising Norwegian reindeer (*Rangifer tarandus tarandus*). *Acta. Physiol. Scand.* **147**, 281-288.
Louw, G. N., Belonje, P. N. and Coetzee, H. J. (1969). Renal function, respiration, heart rate and thermoregulation in the ostrich (*Struthio camelus*). *Scient. Pap. Namib Desert Res. Stn.* **42**, 43-54.
Lund, R. J., Guthrie, A. J., Mostert, H. J., Travers, C. W., Nurton, J. P. and Adamson, D. J. (1996). Effect of three different warm-up regimens on heat balance and oxygen consumption of Thoroughbred horses. *J. Appl. Physiol.* **80**, 2190-2197.
Maina, J. N. and Nathaniel, C. (2001). A qualitative and quantitative study of the lung of an ostrich, *Struthio camelus*. *J. Exp. Biol.* **204**, 2313-2330.
Maloney, S. K., Fuller, A., Mitchell, G. and Mitchell, D. (2001). Rectal temperature measurement results in artefactual evidence of selective brain cooling. *Am. J. Physiol.* **281**, R108-R114.
Maloney, S. K., Fuller, A., Mitchell, G. and Mitchell, D. (2002). Brain and arterial blood temperatures of free-ranging oryx (*Oryx gazella*). *Pflügers Arch.* **443**, 437-445.
Mather, G. W., Nahas, G. G. and Hemingway, A. (1953). Temperature changes of pulmonary blood during exposure to cold. *Am. J. Physiol.* **173**, 390-392.
McFadden, E. R., Jr (1983). Respiratory heat and water exchange: physiological and clinical implications. *J. Appl. Physiol.* **54**, 331-336.
Midtgard, U. (1983). Scaling of the brain and eye cooling system in birds: a morphometric analysis of the rete ophthalmicum. *J. Exp. Biol.* **225**, 197-207.
Mitchell, D., Laburn, H. P., Nijland, M. J. M., Zurovsky, Y. and Mitchell, G. (1987). Selective brain cooling and survival. *S. Afr. J. Sci.* **83**, 598-604.
Mitchell, D., Maloney, S. K., Jessen, C., Laburn, H. P., Kamerman, P. R., Mitchell, G. and Fuller, A. (2002). Adaptive heterothermy and selective brain cooling in arid-zone mammals. *Comp. Biochem. Physiol. B* **131**, 571-585.
Mitchell, D., Maloney, S. K., Laburn, H. P., Knight, M. H. and Jessen, C. (1997). Activity, blood temperature and brain temperature of free-ranging springbok. *J. Comp. Physiol. B* **167**, 335-343.
Pinshow, B., Bernstein, M. H., Lopez, G. E. and Kleinhaus, S. (1982). Regulation of brain temperature in pigeons: effects of corneal convection. *Am. J. Physiol.* **242**, R577-R581.
Refinetti, R. and Menaker, M. (1992). The circadian rhythm of body temperature. *Physiol. Behav.* **51**, 613-637.
Richards, S. A. (1967). Anatomy of the arteries of the head in the domestic fowl. *J. Zool., Lond.* **152**, 221-234.
Richards, S. A. (1970). Brain temperature and the cerebral circulation in the chicken. *Brain Res.* **23**, 265-268.
Richards, S. A. and Sykes, A. H. (1967). Responses of the domestic fowl (*Gallus domesticus*) to occlusion of the cervical arteries and veins. *Comp. Biochem. Physiol.* **21**, 39-50.
Scott, N. R. and van Tienhoven, A. (1971). Simultaneous measurement of hypothalamic and body temperatures and heart rate in poultry. *Trans. Amer. Soc. Agr. Eng.* **14**, 1027-1033.
Simoens, P., Lauwers, H., De Geest, J. P. and De Schaepdrijver, L. (1987). Functional morphology of the cranial Retia mirabilia in the domestic animals. *Schweiz. Arch. Tierheilk.* **129**, 295-307.
Wehr, T. A. (1992). A brain-warming function for REM sleep. *Neurosci. Biobehav. Rev.* **16**, 379-397.
Williams, J. B., Siegfried, W. R., Milton, S. J., Adams, N. J., Dean, W. R. J., du Plessis, M. A., Jackson, S. and Nagy, K. A. (1993). Field metabolism, water requirements, and foraging behaviour of wild ostriches in the Namib. *Ecology* **74**, 390-404.
Withers, P. C. and Crowe, T. M. (1980). Brain temperature fluctuations in helmeted guineafowl under semi-natural conditions. *Condor* **82**, 99-100.