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ORIGINAL PAPER

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Daily energy expenditure of the grey mouse lemur (*Microcebus murinus*): a small primate that uses torpor

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Abstract We aimed to investigate the pattern of utilisation of torpor and its impact on energy budgets in free-living grey mouse lemurs (*Microcebus murinus*), a small nocturnal primate endemic to Madagascar. We measured daily energy expenditure (DEE) and water turnover using doubly labelled water, and we used temperature-sensitive radio collars to measure skin temperature (T_{sk}) and home range. Our results showed that male and female mouse lemurs in the wild enter torpor spontaneously over a wide range of ambient temperatures (T_a) during the dry season, but not during the rainy season. Mouse lemurs remained torpid between 1.7–8.9 h with a daily mean of 3.4 h, and their T_{sk} s fell to a minimum of 18.8 °C. Mean home ranges of mouse lemurs which remained normothermic were similar in the rainy and dry season. During the dry season, the mean home range of mouse lemurs showing daily torpor was significantly smaller than that of animals remaining normothermic. The DEE of *M. murinus* remaining normothermic in the rainy season (122 ± 65.4 kJ day^{-1}) was about the same of that of normothermic mouse lemurs in the dry season (115.5 ± 27.3 kJ day^{-1}). During the dry season, the mean DEE of *M. murinus* that utilised daily torpor was 103.4 ± 32.7 kJ day^{-1} which is not significantly different from the mean DEE of animals remaining normothermic. We found that the DEE of mouse lemurs using daily torpor was significantly correlated with the mean temperature difference between T_{sk} and T_a ($r^2 = 0.37$) and with torpor bout length ($r^2 = 0.46$), while none of these factors explained significant amounts of variation in the DEE of the mouse lemurs remaining normothermic. The mean water flux rate of mouse lemurs using daily torpor (13.0 ± 4.1 ml day^{-1}) was significantly lower than that of mouse lemurs remaining normothermic (19.4 ± 3.8 ml day^{-1}), suggesting the lemurs conserve water by entering torpor. Thus, this first study on the energy budget of free-ranging *M. murinus* demonstrates that torpor may not only reflect its impact on the daily energy demands, but involve wider adaptive implications such as water requirements.

Key words Daily energy expenditure · Doubly labelled water · Grey mouse lemur · *Microcebus murinus* · Torpor

Abbreviations *BMR* basal metabolic rate · *DEE* daily energy expenditure · ΔT mean temperature difference · *FMR* field metabolic rate · *RMR* resting metabolic rate · T_a ambient temperature · T_b body temperature · T_{sk} skin temperature

Introduction

The high mass-specific metabolic rates of small endothermic animals have favoured the evolution of torpor, primarily to minimize energy expenditure, enabling them to cope with periods of cold and food shortage. Torpor is a regulated state of physiological dormancy during which body temperature (T_b) may drop by 4–35 °C below normothermic levels, and metabolic rate may be reduced to as little as 5% of its normothermic value (Geiser and Ruf 1995; Wang 1989 for reviews and references). Because relative heat loss is inversely related to body mass in endotherms (Schmidt-Nielsen 1997), torpor occurs mainly in small mammals, which may be unable to store adequate amounts of energy to

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maintain a constant high T_b during cold exposure (Geiser 1994).

The grey mouse lemur (*Microcebus murinus*), weighing 60 g, is found in the dry deciduous forests in southern and western Madagascar (Mittermeier et al. 1994; Tattersall 1982), and is known for its ability to enter torpor during the cool dry season when temperatures are low and food and water is scarce (Bourlière and Petter-Rousseaux 1966; Martin 1973; Petter-Rousseaux 1975). Before the dry season, seasonal body and tail fattening enable *M. murinus* to effectively face these unfavourable environmental conditions (Fietz 1998; Schmid 1999; Schmid and Kappeler 1998). During their solitary nocturnal activity they feed on fruits, small animals, gum and insect secretions (Corbin and Schmid 1995; Martin 1973). They spend the day alone or in groups in tree holes, and the size of sleeping groups varies seasonally (Pages-Feuillade 1988; Fietz 1995; Radespiel et al. 1998; Schmid 1998). Reproductive activity is photoperiodically controlled and takes place at the beginning of the rainy season (Perret 1992). After 2 months of gestation they give birth to 1–3 young, which are weaned about 2 months later.

Ortmann et al. (1996, 1997) and Schmid (1996, 2000) have measured T_b and oxygen consumption of torpid *M. murinus* kept in outdoor enclosures under natural ambient temperature (T_a) cycles during the dry season in Madagascar. Metabolic rates during daily torpor were reduced to about 20–30% of normothermic mouse lemurs and T_bs dropped to values of less than 10 °C. However, no measurements of torpor and its physiological consequences are available for free-ranging mouse lemurs, nor is anything known about thermoregulation and energy metabolism during the rainy season. The energy budget of an animal can provide much insight knowledge into its physiology, ecology, and evolution (Schmidt-Nielsen 1997), particularly while behaving normally in its natural habitat. Field metabolic rate (FMR) or daily energy expenditure (DEE) is the total energy a wild animal expends during the course of an entire day, which includes the costs of basal metabolic rate (BMR), thermoregulation, locomotion, and other expenses. Energy expenditure and water metabolism in the field can be measured routinely by means of doubly labelled water (Lifson and McClintock 1966; Speakman 1997). We therefore measured DEE, water flux rate and skin temperature (T_{sk}) of free living *M. murinus* in the dry deciduous forest of Kirindy/CFPF in Madagascar during both the rainy and dry seasons. The main questions addressed in this study were:

1. Is there a seasonal difference in the daily energy expenditures of mouse lemurs?
2. How much energy is saved by using torpor?
3. How are T_as, torpid T_bs, and torpor bout durations related to the DEE of mouse lemurs using daily torpor?

Materials and methods

Study site

We conducted the study in three primary forest plots (CS7, N5 and CS5) in the forestry concession, the Centre de Formation Professionelle Forestière de Morondava (CFPF). Each plot is equipped with a regular trail system consisting of small foot trails every 25 m in both north-south and east-west directions, and each intersection is marked with a plastic tag for orientation. The plots are part of a ca. 12,000-ha tract of deciduous dry forest in western Madagascar (Forêt de Kirindy/CFPF) 60 km northeast of Morondava (44°39′E, 20°03′S). General information on the phenology and history of this forest is given by Ganzhorn and Rohner (1996). The area around Morondava has pronounced seasonality with a hot rainy season between December and March and a cool dry season of 7–8 months with virtually no precipitation. The mean temperature during the rainy season is 27.4 °C, with mean maxima and minima of 27.0 °C and 27.4 °C, respectively. Temperature during the dry season shows extreme diurnal variation, with cold nights (4 °C) and hot days (32 °C), and a mean of 23.5 °C (Sorg and Rohner 1996). Measurements were performed during the rainy season from December 1997 until January 1998, and during the dry season from April until June 1998.

Trapping

Sherman live traps (7.7 × 7.7 × 30.5 cm) were baited with small pieces of banana and set at each 25-m junction of the study grid system, 1–2 m above the ground. On experimental days, traps were opened and baited at dusk and checked and closed at dawn. All mouse lemurs captured were taken to the research station (between 0.5 km and 1.5 km from the trapping areas) for identification and measurements. Mouse lemurs were weighed using a 100-g spring balance to the nearest 0.5 g, sexed, examined for reproductive state, and on the first capture were individually marked by subdermal injection of a transponder (Trovan, Weilerswist, Germany).

Telemetry

We used radio collars (SS-2 button cell tags, accuracy ± 1 °C) with attached temperature sensitive sensors (Biotrack, Wareham, Dorset, UK) to monitor physiological state, location and movement of the animals . Transmitters weighed on average 2.5 g which is below the tolerable weight limit of 5% of the animals body mass (Gursky 1998). Prior to attachment, transmitters were calibrated against a mercury thermometer in a water bath between 4 °C and 40 °C (± 0.5 °C). The radio collars were placed so that the temperature sensor was in contact with the animal's skin on the ventral part of the neck. This T_{sk} is a reasonable estimate of core body temperature (Audet and Thomas 1996) particularly when resting or torpid animals are in a curled-up position with the transmitter pressed against the ventral surface. After release we took manual transmitter readings using a TR-4 receiver (Telonics, Mesa, Ariz., USA) protected by a waterproof environmental housing (Sexton Photographics, Salem, Ore., USA) with a flexible two-element Yagi antenna and a stopwatch. The effective range of signal reception (frequency 150.000–151.000 MHz) varied from 50 m to 150 m depending on the terrain.

During the rainy season, each animal was followed for 6 h per night (1800–2400 hours). During the dry season, we followed the animals until they ceased activity and either entered torpor or remained inactive in their daytime sleeping sites. Positions of the mouse lemurs were taken approximately every 30 min and determined by two bearings taken from two different intersections of the defined trail system. Locations of the animals were triangulated with the computer software Track3 (A. Ganzhorn, 1993 unpublished observations). Home ranges were analysed with Tracker (Camponotus AB, Solna, Sweden) using the minimal convex polygon technique. Parallel to each position reading, we determined T_{sk} by averaging three timings of the interval required for ten transmitter pulses and then using the calibration curves prepared for each transmitter. After the activity period all radio-collared animals were located in their sleeping sites. To monitor physiological state over the day, we took transmitter readings as described above approximately every 3 h until mouse lemurs restarted their activity at dusk (approximately 0700–1900 hours). To recapture animals investigated, we set 15–20 traps located within a radius of 50 m of the sleeping site. If mouse lemurs were not recaptured within the first night after radio tracking, mobile daytime recordings of T_{sk} were repeated until the animals were recaptured (up to 4 successive days). We did not take transmitter readings when mouse lemurs were active because trapping success may be affected by following them. At the same time that we measured T_{sk}, we used a digital thermometer (GTH 1200; Geisinger Electronic) at the tracking position to measure T_a to the nearest 0.5 °C.

Daily energy expenditure

Mouse lemurs used for measurement of DEE included males and females which were not reproductively active. For males, reproductive inactivity refers to individuals with small but visibly developed testes. For females, nonreproductive status refers to individuals which did not show signs of pregnancy (palpating) and lactation (swollen teats). Each individual was only used once in the field measurement period, either during the rainy or dry seasons. Animals selected for measurement were injected intraperitoneally with 0.4 ml water containing $H_2{}^{18}O$ (10 APE) and D_2O (6 APE). This dose was anticipated to be sufficient for a measurement lasting between 2 days and 5 days (Speakman 1997). After injection the mouse lemurs were held in Sherman traps for at least 1 h to allow complete equilibration of the isotopes in the body water. Animals were briefly (approx. 10–20 min) anaesthetised with 0.03 ml Ketavet (100 mg ml^{-1}) and an initial blood sample was taken by vena puncture of the ear veins. Body mass was measured and the animal was fitted with the radio-collar. Upon recapture, between 2 days and 3 days later, body mass was determined again, a second blood sample was taken and the radio collar was taken off. After participation in the measurement, animals were released at their capture site on the same day in the late afternoon. Blood samples were flame sealed into 50 μ-l capillaries and transported to Aberdeen (UK) for analysis.

Blood samples were distilled using the pipette method of Nagy (1983). Mass spectrometric analysis of deuterium enrichment was performed using H_2 gas, produced from the distilled water after reaction with $LiAlH_4$ (Ward et al. 2000). For analysis of oxygen-18 enrichment, distilled water was equilibrated with CO_2 gas using the small sample equilibration technique (Speakman et al. 1990). ^2H:^1H and ^{18}O:^{16}O ratios were measured using dual inlet gas source isotope ratio mass spectrometry (Optima, Micromass IRMS), with isotopically characterised gases of H_2 and CO_2 (CP grade gases BOC Ltd) in the reference channels. Reference gases were characterised every 3 months relative to SMOW and SLAP (Craig 1961) supplied by IAEA. Each batch of samples was run with triplicates of three laboratory standards to correct for day-to-day variation in performance of the mass spectrometers. All isotope enrichments were measured in delta (per millilitre) relative to the working standards and converted to parts per million, using the established ratios for these reference materials. Measures of isotope enrichment were based on independent analysis of two sub-samples of the water distilled from the blood samples.

We estimated CO_2 production using the single pool deuterium equation (7.17) from Speakman (1997) as recommended by Visser and Schekermann (1999). The error in individual estimates was determined using the iterative procedures outlined in Speakman (1995). Conversion to energy expenditure was made using an assumed RQ of 0.8. All calculations were made using the Natureware DLW software (Speakman and Lemen 1999).

A total of eight males and two females during the rainy season, and 20 males and 20 females during the dry season, respectively, were injected with $D_2{}^{18}O$ and successfully recaptured. However, blood samples of 20 animals were so small that the analyses of D_2 and ^{18}O could not be performed. Thus, final sample size for measurements of the daily energy expenditure was reduced to six males and one female in the rainy season, and to 11 males and 12 females in the dry season.

Data analyses

For each individual, a mean T_{sk} when the mouse lemurs remained normothermic was calculated for each the resting phase (0600–1800 hours) and the activity phase (1800–0600 hours). We also averaged corresponding T_a readings for each phase. For mouse lemurs that were found torpid in the traps, we determined the onset of torpor by using the calculated cooling rate for this species of 3.4 °C h^{-1} (Schmid 2000). Mouse lemurs were considered to be torpid when the T_{sk} was equal to or lower than 30 °C for at least 1 h, i.e. at least two temperature readings (Geiser and Baudinette 1988). The total length of each torpor bout was divided into two different phases: (a) the length of torpor during the resting phase, and (b) the length of torpor during the activity phase. For each torpor episode, parameters analysed included: (1) the total length of each torpor bout in hours, defined as the time between torpor onset when the T_{sk} fell below 30 °C and the end of the torpor bout defined by the first measurement when T_{sk} reached normothermic levels again, (2) duration of torpor during the resting phase and activity phase, respectively, in hours, (3) mean T_{sk} when the body temperature was at the minimum point during each torpor bout, and (4) mean of the corresponding T_a during torpor. For animals that showed two or more torpor episodes within the period of time of the DEE measurement, we calculated a single average for each parameter.

All results are given as mean ± SD. Differences between means were examined using Student's-*t*-test and analysis of variance (ANOVA). The Pearson correlation coefficient and linear regression coefficient were calculated to test for significant correlations between variables. Torpor bout length was log-transformed for statistical analyses. All statistical analyses were performed following Sokal and Rohlf (1981) using StatView 4.02.

Results

Thermoregulatory status

During the rainy season, mouse lemurs remained completely normothermic within periods of measurements, and torpor was not observed. For the whole set of 20 males and 20 females examined in the dry season, 9 individuals of each gender entered torpor, and 11 remained normothermic over the measurement period. Figure 1 shows typical patterns of change in T_{sk} over an entire 2-day tracking period (20–22 May) for an animal which remained normothermic, and one which entered daily torpor. Separated by season and thermoregulatory status, mean T_a and T_{sk} of animals are given in Table 1. Across all 50 mouse lemurs, the mean T_{sk} (36.5 ± 0.9 °C) during the active phase was significantly higher than the mean T_{sk} (35.5 ± 1.5 °C) for the resting phase ($t = 4.34$, $P < 0.0001$). The overall mean T_a was 22.1 ± 2.4 °C for the active phase, and 22.6 ± 5.5 °C for the resting phase, which was not significantly different. However, fluctuations of T_a were greater, and T_a was lower in the dry season than in the rainy season. The individual minimum T_{sk} in torpor was 18.8 °C, which we

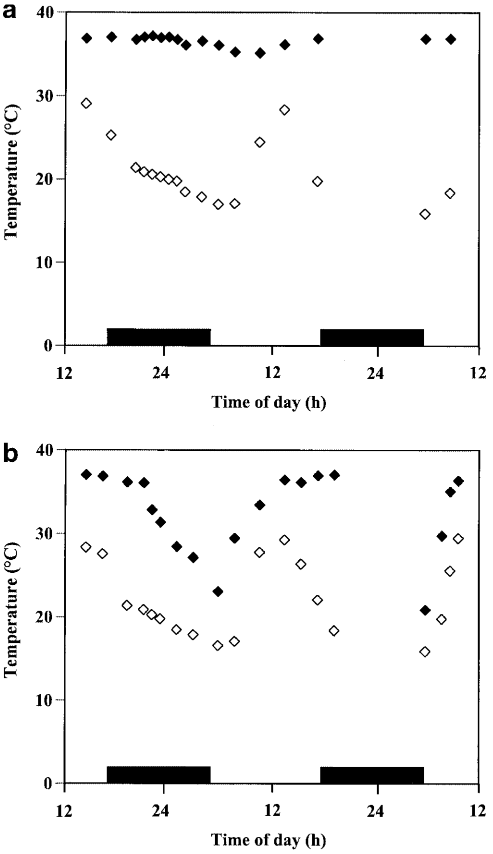

Fig. 1 Registration of body temperature (*filled symbols*) and ambient temperature (*open symbols*) of two *Microcebus murinus* during **a** normothermia (animal M59), and **b** torpor (animal M60) recorded on May 20–22 1998 during the dry season

Table 1 Temperature characteristics for *Microcebus murinus* during the rainy and dry season 1997/1998. Values are means ± standard deviations of the total set of animals investigated. (T_a ambient temperature, T_b body temperature, T_{sk} skin temperature)

	Rainy season Normothermic (n = 10)	Dry season	
		Normothermic (n = 22)	Torpid (n = 18)
T_{sk} active (°C)	36.7 ± 0.8	36.7 ± 1.0	36.1 ± 0.9
T_{sk} resting (°C)	36.1 ± 0.9	35.7 ± 1.7	34.7 ± 1.2
Minimum T_{sk} torpor (°C)			24.1 ± 2.7
T_{sk} mean (°C)	36.4 ± 0.6	36.2 ± 1.2	32.6 ± 0.7
T_a active (°C)	29.4 ± 1.1	20.6 ± 2.1	19.9 ± 2.6
T_a resting (°C)	29.4 ± 2.6	19.2 ± 3.3	22.9 ± 5.4
T_a torpor (°C)			16.5 ± 3.5
T_a mean (°C)	29.4 ± 1.6	19.9 ± 2.3	19.8 ± 3.6
(T_{sk}−T_a) mean (°C)	7.0 ± 1.9	16.3 ± 2.2	12.3 ± 3.4

recorded for male M69 in May at a T_a of 12.9 °C. The highest individual T_{sk} during a torpor bout was 29.0 °C measured in female F52 in May at a T_a of 22.0 °C.

For all animals investigated, the mean duration of daily torpor bout was 3.4 ± 1.7 h ($n = 18$). The longest individual daily torpor bout recorded was 8.9 h (animal F52) and the shortest was 1.7 h (M68 and F64), respectively.

Home range

When the subset of mouse lemurs remaining normothermic is considered, the mean home range during the rainy season ($n = 10$, 2562 ± 1959 m^2) did not differ significantly from that during the dry season (mean: $n = 22$, 2203 ± 1218 m^2; $t = 0.58$, $P > 0.5$). During the dry season, the mean home range of mouse lemurs which used torpor ($n = 18$, 1408 ± 958 m^2) was significantly smaller than the mean home range of mouse lemurs which remained normothermic ($t = 2.33$, $n = 22$, $P = 0.03$). There was a significant negative correlation between home range and torpor duration in mouse lemurs using daily torpor ($r = -0.52$, $n = 18$, $P = 0.02$).

DEE in free-living mouse lemurs

Background isotope levels in free-living mouse lemurs averaged 166.6 ± 12.9 ppm for deuterium ($n = 12$) and 2008.9 ± 7.3 ppm for ^{18}O ($n = 8$). The individual-to-individual variation in isotope abundance for ^{18}O ranged from 2000.3 ppm to 2019.9 ppm, and from 147.6 ppm to 187.0 ppm for hydrogen.

For the following analysis only animals for which DEE was determined were taken into account ($n = 30$). The period of time between the first and the second blood sample averaged 2.3 ± 0.5 days (range 1.7–3.8 days). During the rainy season, DEE of *M. murinus* averaged 122 ± 65.4 kJ day^{-1}, which was 2.4-times their estimated resting metabolic rate (RMR; data from Perret et al. 1998; Table 2). Individual DEE values ranged from 0.6- to 4.1-times the resting metabolism. One animal had a DEE that was lower than its predicted resting rate which we can not explain, but probably reflected an error in the isotopic analysis. In the seven normothermic mouse lemurs, the mean T_{sk} was 36.9 ± 0.6 °C, and the corresponding mean T_a was 29.2 ± 1.4 °C.

During the cool and dry months from April until June 1998, we measured both T_{sk} and DEE values from 11 animals that remained completely normothermic, and from 12 individuals that used torpor during parts of the field measurement period. The mean DEE of mouse lemurs that were normothermic ($n = 11$) was 115.5 ± 27.3 kJ day^{-1}, and their mean T_{sk} was 35.8 ± 1.2 °C at a mean T_a of 19.1 ± 3.1 °C (Table 3). The ratio between DEE and estimated resting metabolic rate ranged from 1.6 to 4.4 and was on average 3.2 ± 0.8. For the sub-sample of mouse lemurs which remained normothermic in the rainy and dry season ($n = 18$), comparisons between the seasons revealed no significant differences in the mean DEE ($t = -0.28$, $P > 0.7$) and the mean T_{sk} ($t = -1.19$, $P > 0.2$). In contrast, the mean temperature difference between T_{sk} and T_a (ΔT) in

Table 2 Summary of daily energy expenditure (DEE) in *Microcebus murinus* in Western Madagascar during the rainy and dry season 1997/1998. (*M* = male, *F* = female). Resting metabolic rate (RMR) was calculated using following regressions for mouse lemurs (Perret et al. 1998): *rainy season*: RMR (ml O_2 h^{-1}) = 74.6 + 0.51 * body mass (g); *dry season*: RMR (ml O_2 h^{-1}) = 30.9 + 0.68 * body mass (g). The resulting values were converted into units of energy equivalence (kJ day^{-1}) assuming the factor 20.083 kJ/l O_2 (Schmidt-Nielsen 1997)

Animal no.	Mean mass (g)	Torpor (h day^{-1})	Daily energy expenditure		Water flux rate (ml day^{-1})
			KJ day^{-1}	Relative to RMR	
Rainy season					
M4	56.0	0	94.34	1.90	13.68
M5	55.0	0	28.46	0.58	17.28
M7	63.0	0	103.03	2.00	12.96
M23	63.8	0	207.35	4.02	19.68
M28	63.0	0	210.11	4.08	18.48
M29	54.3	0	97.08	1.97	22.80
F6	67.0	0	111.03	2.12	23.04
Mean ± SD	60.3 ± 5.1	0	121.63 ± 65.40	2.38 ± 1.26	18.27 ± 3.99
Dry season					
M30	67.0	0	60.02	1.64	23.04
M36	60.0	0	138.37	4.02	22.32
M59	57.0	0	103.81	3.02	22.32
M71	70.0	0	129.13	3.40	16.08
M73	63.0	0	118.94	3.41	13.68
Mean ± SD	63.3 ± 4.8	0	110.05 ± 30.77	3.10 ± 0.89	19.49 ± 4.30
M60	61.0	2.4	166.50	4.77	4.08
M61	59.0	3.3	85.37	2.49	12.96
M62	43.5	2.2	86.78	2.98	11.52
M68	55.5	1.7	149.40	4.53	13.68
M70	38.5	2.6	86.50	3.14	13.20
M72	54.0	3.1	120.37	3.69	10.80
Mean ± SD	51.9 ± 8.9	2.6 ± 0.6	115.82 ± 35.63	3.60 ± 0.9	11.04 ± 3.58
F34	98.0	0	150.67	3.20	19.44
F40	66.5	0	111.94	3.05	18.48
F44	57.0	0	102.21	3.04	26.88
F57	72.0	0	81.45	2.12	16.56
F63	72.5	0	139.49	3.61	18.72
F66	48.5	0	134.46	4.37	15.84
Mean ± SD	69.1 ± 16.9	0	120.04 ± 26.07	3.23 ± 0.74	19.32 ± 3.95
F31	47.0	2.5	97.33	2.69	15.36
F32	86.0	3.1	88.83	3.14	20.64
F52	97.5	8.9	50.49	1.76	10.08
F56	84.5	3.1	75.02	1.08	12.24
F64	79.0	1.7	128.25	2.06	17.52
F65	75.0	2.5	106.02	3.21	13.68
Mean ± SD	78.2 ± 17.1	3.6 ± 2.6	90.99 ± 26.66	2.32 ± 0.84	14.92 ± 3.79

Table 3 Temperature characteristics for *M. murinus* remaining normothermic and entering torpor, respectively. Only the subset of animals investigated during the dry season for which DEE was determined were taken into account. Values are means ± SD, minima, and maxima (given in parentheses). Student's-*t*-test was used for statistical comparisons between normothermia and torpor for each parameter (*ns* not significant)

	Normothermic (n = 11)	Torpid (n = 12)	*t*-test
T_{sk} mean (°C)	35.8 ± 1.2 (33.3/37.2)	32.6 ± 0.7 (31.0/33.3)	$t = 7.98$ $P < 0.0001$
T_a mean (°C)	19.1 ± 3.1 (14.0/23.4)	19.1 ± 3.6 (13.6/25.2)	ns
(T_{sk}–T_a) mean (°C)	16.5 ± 3.0 (11.8/21.6)	12.9 ± 3.6 (6.5/18.7)	$t = 2.65$ $P = 0.02$
Minimum T_{sk} torpor (°C)		23.9 ± 2.6 (20.5/28.3)	

the dry season (16.5 ± 3.0 °C) was significantly greater than that in the rainy season (7.1 ± 1.8 °C; $t = 7.51$, $P < 0.0001$). ΔT had no significant effect on DEE of mouse lemurs remaining normothermic in both seasons ($r^2 = 0.03$, $n = 18$, $P = 0.5$).

DEEs of mouse lemurs that utilised daily torpor were variable, ranging from 50.5 kJ day^{-1} to 166.5 kJ day^{-1} (Table 2). During the dry season, the mean DEE of mouse lemurs using torpor did not differ significantly from the mean DEE of animals remaining normothermic

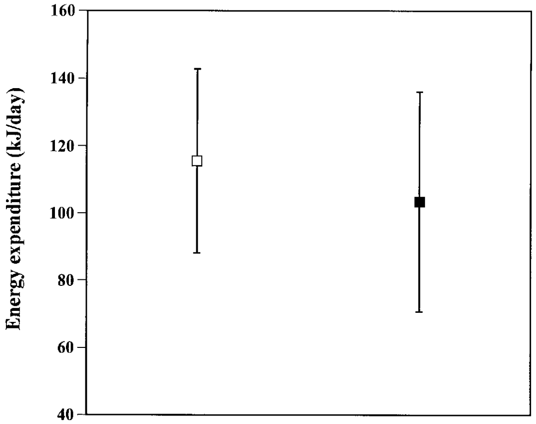

Fig. 2 Daily energy expenditure during torpor (*filled symbols*; $n = 12$) in comparison to normothermia (*open symbols*; $n = 11$) in *M. murinus* during the dry season. Values represent means ± SD

($t = 0.96$, $n = 23$, $P = 0.35$) (Fig. 2). Separated by sex, however, the average DEE of females using torpor was reduced by 24.2% compared to the mean DEE of females remaining normothermic, whereas the mean DEE of males showing torpor did not differ from that of males remaining normothermic (mean values see Table 2). For the subset of mouse lemurs investigated during the dry season for which DEE was determined, means of temperature characteristics (T_{sk}, T_a, ΔT, min T_{sk} torpor) are given in Table 3. We found that DEE of mouse lemurs using daily torpor ($n = 12$) was significantly correlated with ΔT ($r^2 = 0.37$, $P = 0.035$) and torpor bout length ($r^2 = 0.46$, $P = 0.015$; Fig. 3).

Water flux rate

Water flux rates were highly variable between animals of different thermoregulatory status (Table 2). During the dry season, mean water flux rate in mouse lemurs using daily torpor were significantly lower than in mouse lemurs remaining normothermic ($t = 3.86$, $P < 0.001$; Fig. 4), suggesting the lemurs conserve water by entering torpor. However, torpor bout length did not explain a significant amount of variance in water flux rates ($r^2 = 0.05$, $n = 12$, $P = 0.48$).

Discussion

The purpose of this study was to determine the patterns of utilisation of torpor during the rainy and dry season, and to quantify the effect of torpor on the DEE. We found that adult free-ranging grey mouse lemurs of both sexes enter daily torpor over a wide range of T_as during the dry season, but not during the rainy season. Since in the present study T_as during the rainy season never decreased for a longer period of time to values below 25 °C, the lemurs could hardly display torpor with body temperatures below 30 °C. It remains unclear, however,

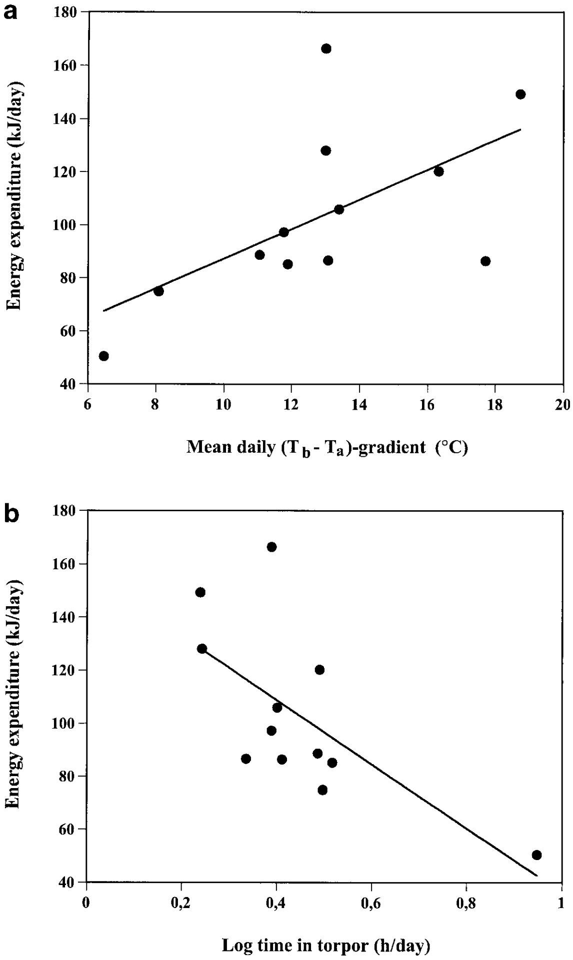

Fig. 3 Daily energy expenditure of mouse lemurs using daily torpor during the dry season ($n = 12$) as a function of the mean skin temperature minus ambient temperature gradient ($T_{sk}-T_a$) **a**, and torpor bout length (torpor bout length is log 10 transformed) **b**. The calculated linear regression for the gradient-induced increase of energy expenditure is DEE = 31.28 + 5.60* ($T_{sk}-T_a$); $r^2 = 0.37$, $P = 0.03$. DEE increases with increasing duration of torpor bouts (linear regression: DEE = 157.28–121.03 *log torpor bout length; $r^2 = 0.46$, $P = 0.015$)

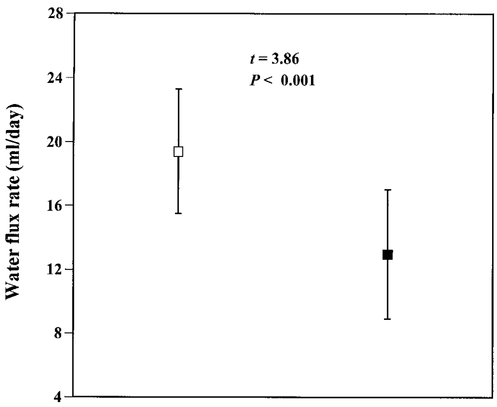

Fig. 4 Water flux rate during daily torpor (*filled symbols*; $n = 12$) in comparison to normothermia (*open symbols*; $n = 11$) in *Microcebus murinus* during the dry season. Values represent means ± SD; Student's-*t*-test was used for statistical comparison

if *M. murinus* would in fact utilise daily torpor in response to unpredictable low temperatures in the rainy season. This aspect of the use of torpor requires further long-term investigations.

Normothermic mouse lemurs regulated T_{sk} over a wide range of T_a between about 32 °C and 38 °C when at rest, but increased their T_{sk} significantly during nocturnal activity. This regulation of T_b has already been reported for this species (Aujard et al. 1998; Ortmann et al. 1997; Schmid 1996, 2000) and is similar to reports for other prosimians (Dobler 1978; McCormick 1981; Müller and Jaschke 1980; Schmid and Ganzhorn 1996; Schmid et al. 2000). The total DEEs of males and females which remained normothermic were similar in the rainy and dry season. However, if the very low DEE of animal M5 (Table 2) is excluded, on the basis it was physiologically not understandable and probably reflected an error in the isotope analysis, energy expenditure during the rainy season averaged 137.2 ± 55.7 kJ day^{-1} ($n = 6$), which was 19% higher than the mean DEE recorded for normothermic animals in the dry season. This difference is surprising, because T_a was lower in the dry season and thus thermoregulatory costs would be expected to be higher. Moreover, since we selected reproductively inactive individuals this difference cannot reflect the additional energy requirements of reproduction (Kenagy et al. 1989; Speakman and Racey 1989). Previous laboratory studies indicated *M. murinus* had a significantly reduced RMR at thermoneutrality when exposed to short daylength (equal to dry season; Chevillard 1976; Perret et al. 1998). Thus, the metabolic savings attributable to the low RMR during the dry season may be sufficient to compensate for the increased thermoregulatory demands of remaining normothermic.

The DEE of normothermic *M. murinus* in the present study was 2.4-times the RMR during the rainy season, and about 3.2-times the resting metabolism during the dry season, respectively. The values are close to the mean value of 3.4 (FMR:RMR) calculated by Speakman (2000) based on 60 small mammalian species weighing less than 4 kg, as well as the average ratio of FMR to RMR of 3.1 measured for the red-tailed sportive lemur *Lepilemur ruficaudatus* (Drack et al. 1999). Grey mouse lemurs which remained normothermic had field metabolic rates that averaged 120% of those expected for eutherians having the same body mass (Nagy 1987: eq. 18). Previously, the grey mouse lemur has been described as having a BMR of only 70% of the value expected for the BMR of eutherian mammals of its body mass (Müller 1985; Ross 1992). However, recent studies have shown that RMR of normothermic *M. murinus* during the breeding season (equals rainy season) averages about 40% above, and during the resting season (equals dry season) 20% below the value expected from the allometric equation of McNab (1988). The results of the present study are clearly inconsistent with the suggestion that DEE might be set at a constant multiple of RMR (Koteja 1991; Peterson et al. 1990; Ricklefs et al. 1996; Weiner 1992).

It has been argued that the costs of locomotion is an important factor influencing the energy budget and that low costs of travel are maintained by very short nightly travel distances (Warren and Crompton 1998). In particular, this might be very important to *M. murinus* since they are under energetic stress during the cool dry season (Martin 1973). The data at hand support this argument. Mouse lemurs using daily torpor had smaller home ranges and therefore maintained low costs of locomotion compared to animals remaining normothermic. These data are consistent with the findings that mouse lemurs housed in outdoor enclosures in Madagascar showed a significant reduction of activity on days with torpor (Ortmann et al. 1997).

DEE reflects the sum of metabolic rates during activity and rest and, if the animal utilises torpor, the metabolic rate during a torpor episode (Karasov 1981). Based on our physiological data, we found that DEE of mouse lemurs using daily torpor was negatively related to both the differential between T_{sk} and T_a and torpor bout length, suggesting that torpor has an impact on the daily energy budget. These observations in mouse lemurs are consistent with the prediction that levels of energy expenditure should be linked to duration of torpor and T_b during torpor (Bartels et al. 1998; Geiser and Kenagy 1988). Nonetheless, we found no significant difference between the mean DEE of *M. murinus* that used daily torpor and that of animals remaining normothermic. This is rather surprising since in *M. murinus* the metabolic rate during torpor is reduced by almost 80% compared to the resting metabolic rate (Schmid 2000). At least two explanations for this discrepancy are possible. First, mouse lemurs in this study remained in torpor for, on average, only 3.4 h, suggesting that the reduced metabolic rate during torpor was not sufficient to account for a significant decrease of the daily energy budget. In addition, one would assume that the energy saved by a short torpor bout should be comparatively low because the reduced torpid metabolic rate cannot compensate for the high energetic costs of arousal. However, this is not the case for mouse lemurs because the costs of arousal are reduced due to the exogenous heating of T_b along with the daily heating phase of air temperature without a noticeable increase in metabolic rate (Schmid 1996, 2000). Furthermore, in the present study the mean minimum T_{sk} during torpor was 23.9 °C and energy expenditure was therefore not reduced substantially to make a significant impact on the total energy budget. Second, the fact that the average DEE of *M. murinus* using torpor versus animals remaining normothermic did not differ does not mean that, on an individual basis, torpor did not have an effect. It is indisputable that mouse lemurs must have saved energy due to the reduced metabolic rate during torpor. However, they may have used up these energy savings for other additional energy intensive activities or processes during the period when they were normothermic, so that the total energy expenditure was not reduced. In other words, if torpor had not occurred, the DEE of those individuals would have been considerably higher. Such intraspecific variations in the daily energy budget has been reported in several small mammals (Koteja 1991; Nagy 1987 for review and references) as well as in the Red-tailed Sportive Lemur (Drack et al. 1999).

Although there was no evidence of a significant reduction of the mean DEE of mouse lemurs using daily torpor compared to that of animals remaining normothermic, the mean DEE of torpid females was reduced by 25% compared to normothermic females (Table 2). This observed reduction comes close to the energy savings we would expect according to the calculated daily energy saving of 37.7% resulting from hypothermia in *M. murinus* kept under natural T_a cycles (Schmid 2000). In contrast, in males that used torpor, the mean DEE was about the same as that of males remaining normothermic. However, the question as to whether the use of daily torpor in *M. murinus* and its function is in fact gender dependent remains speculative, and to describe crucial factors determining the use of torpor and its energetic consequences requires long-term recordings of torpor patterns and energy budgets in free-ranging animals.

Mean values of water turnover in *M. murinus* were significantly lower in animals that entered torpor compared to animals that remained normothermic. These low rates of water loss were presumably associated with low ventilation rates required for low metabolic rates and a low T_b. For mouse lemurs in this study it therefore appears that the main benefit of torpor was that it conserved water loss and possibly thereby reduced water requirements. Since there is virtually no precipitation during the dry season around Morondova (Sorg and Rohner 1996), the savings in their water budgets may be a significant factor influencing the utilisation of torpor. Variation in the use of torpor by individual mouse lemurs could thus reflect essentially stochastic processes relating to their discovery of water during each activity period. Torpor in response to water restriction or deprivation has been documented for several other small mammals (Song and Geiser 1997; Thomas and Cloutier 1992).

In summary, our study suggests that the function of torpor may be different in different individuals within the same species and may not only reflect its impact on daily energy demands, but involve much wider adaptive implications, perhaps including water requirements. An important conclusion from this work is that while daily torpor involves very low energy expenditures this need not necessarily lead to a significant reduction of the daily energy budget.

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