# Effects of ambient temperature on tropical hibernation in the lesser hedgehog tenrec, *Echinops telfairi*

## Dissertation

zur Erlangung des Doktorgrades (Dr. rer. nat.)
des Departments Biologie der Fakultät für Mathematik, Informatik
und Naturwissenschaften an der Universität Hamburg

vorgelegt von

**Jens Wein**

Hamburg, Juni 2010

Genehmigt vom Department Biologie
der Fakultät für Mathematik, Informatik und Naturwissenschaften
an der Universität Hamburg
auf Antrag von Frau Professor Dr. K. DAUSMANN
Weiterer Gutachter der Dissertation:
Professor Dr. W. J. FOLEY
Tag der Disputation: 13. August 2010

Hamburg, den 29. Juli 2010

64

A. Temming
Professor Dr. Axel Temming
Leiter des Departments Biologie

THE AUSTRALIAN NATIONAL UNIVERSITY

Research School of Biology
Division of Evolution, Ecology & Genetics
Professor William J. Foley
Professor of Zoology and Head of Division

CANBERRA ACT 0200 AUSTRALIA

TELEPHONE: +61 26 125 2866
FACSIMILE: +61 26 125 5573
EMAIL: william.foley@anu.edu.au

May 29 2010

**To Whom it May Concern**

As a native speaker I hereby confirm that the PhD thesis of Jens Wein, titled "Effects of ambient temperature on tropical hibernation in the lesser hedgehog tenrec, *Echinops telfairi*" is written in good English grammar and comprehensible style.

Yours sincerely.

William J Foley
**Professor and Head, Division of Evolution, Ecology and Genetics**

*I have gotten a lot of results. I know several thousand things that won't work.*

Thomas A. Edison

# Contents

# Chapter 1

# Introduction

## 1.1 Energy expenditure

Of all fundamental principles in biology, energy expenditure is perhaps the single-most important as it is a prerequisite for life. All organisms on this planet have to consume energy in order to stay alive. In contrast to plants and other photoautotrophic organisms that utilize solar energy to fuel their metabolism, animals rely on energy-rich molecules supplied in their food[1]. Since food is limited and high-energy food is even scarcer, food becomes, besides oxygen and water, the most important resource for survival. Precisely because energy plays such a critical role in animal survival, numerous strategies have evolved to maximize energy intake and to minimize energy expenditure. Because the amount of energy that can be assimilated is ultimately limited by the energy supplied by the habitat, it seems more feasible to achieve an economic energy budget by reducing energy consumption.

## 1.2 Composition of energy consumption

Mammals, like all endotherms, are able to maintain a constant high body temperature ($T_b$) by endogenous heat production. A constant high $T_b$ makes mammals highly efficient in numerous ways and allows them to conquer habitats inaccessible to ectotherms. It is even believed that the ability to maintain a constant high $T_b$ is a major reason for their evolutionary success (Miller and Harley, 1996). But this ability comes at a high cost, namely high energy requirements. When compared to ectotherms of equal body size and at the same $T_b$, mammals expend around eight times more energy (Else and Hulbert, 1981). Even at the same $T_b$ and within the thermoneutral zone, mammalian metabolism always requires more energy.

---

[1] An exception to this are animals that lead a symbiotic lifestyle with photo- or chemoautotrophic symbionts, e.g. stony corals and zooxanthellae or beard worms and sufur-reducing bacteria.

Mammalian metabolic rate (MR) consists of various processes that account for different, but dynamic proportions of overall energy expenditure. The MR of a fasted adult animal at rest in its thermoneutral zone is called basal metabolic rate (BMR)[2]. Basal metabolic rate comprises all processes that are essential to keep the "machinery" of the animal running. This includes vital processes such as respiratory, circulatory and renal activity. But other maintenance processes, e.g. preservation of electro-chemical gradients by ion pumps or RNA and protein biosynthesis add to BMR as well.

Not surprisingly, overall, large animals need more energy than do small animals. But the relation between body mass and BMR is not linear. If MR is calculated per unit body mass, bigger animals need less energy than do smaller ones. This allometric scaling of BMR in the form of $MR = a \cdot M^b$ [3] was first investigated in detail by Kleiber (1932). He proposed that BMR scaled to body mass with an exponent of 3/4 rather than to the exponent of 2/3 as previously proposed, yielding $BMR = 3.9 \cdot M^{0.75}$ [4]. To date numerous studies have been undertaken that have compared BMR to body mass in various clades of mammals and birds, as well as ectotherms and even single-cell organisms. A comparison across all mammals, including more than 250 species, revealed a body mass exponent of 0.71 rather than the exponent of 0.75 (Heldmaier, 2003; Lovegrove, 2003). The exponent seems to be quite stable when compared across systematic groups, but can vary widely within different mammalian orders and exponents from 0.42 to 1.08 have been observed (Hulbert and Else, 2000).

The reason why BMR does not scale linearly with body mass is still under debate. The MR of smaller mammals is higher when thermoregulatory costs are included. Small animals have a high surface to volume ratio and thus lose more heat to their surroundings than larger animals, especially in cold conditions. But why this also affects BMR, which is measured in the thermoneutral zone and therefore excludes thermoregulatory costs, is less clear. Evolutionary, there is a tendency to increase MR proportionally to body mass (b=1) since larger animals are copies of small animals, but the geometry of body structures means that surface area changes only with the two third power of size (b=0.67), and this produces a fundamental conflict (Hemmingsen, 1960). Surface-volume ratio is certainly of major importance. Absorption of food or respiratory gases and waste elimination are all functions related to surface area and so limit metabolism with increasing size. With this effect alone one would obtain an exponent of 0.67 because volume increases to the power of three while surface area only increases to the power of two with increasing body mass. It has been suggested that other aspects might also play a role, such as changing structural demands on the skeletal system and metabolising organs with increasing body size (McMahon, 1973) or structural and functional properties of vertebrate cardiovascular and respiratory systems (West et al., 1997). Hulbert and Else (2000) argued that species would be selected for decreases in their mass-specific MR or for increases in the areas of the surfaces

---

[2] since the requirement of a fasted animal at rest is sometimes impracticable to satisfy, the MR during a stretch of time where the animal is calm is taken and named resting metabolic rate (RMR)

[3] this formula describes overall energy consumption with M being body mass (g)

[4] when noted per gram body weight, the formula is $BMR = 3.9 \cdot M^{-0.25}$

that service metabolism as they increased in body size. Because both evolutionary strategies were followed, the measured exponents represent a compromise between these two tendencies.

Although BMR is a valuable tool for interspecific or phylogenetic comparisons on a larger scale, its relevance for ecological and evolutionary aspects of a species is arguable (Kozlwoski and Weiner, 1997; Kozlowski and Konarzewski, 2005). Every activity or process in an animal that goes beyond maintenance of the status quo requires further energy. Locomotion, growth, mating, communication or thermoregulation will increase MR, to name just a few. From this it follows that daily energy expenditure (DEE) will exceed BMR many fold. Captive animals have a DEE of about 2-3 times the BMR and the MR of free-ranging animals in the wild are 3-4 times the BMR (Heldmaier, 2003). Therefore, the forces of natural selection will act on DEE rather than BMR. In order to gain an advantage over competing species, evolution has, and will, favour species that use energy efficiently.

## 1.3 Adaptations for energy conservation

In order to save energy, evolution led to many different adaptations in mammals. Since energy saving is most important in cold climates, where the temperature difference between the animal and its surrounding is most pronounced, it is not surprising that the most sophisticated adaptations and mechanisms are found in animals from temperate and arctic regions.

The adaptations employed may affect the animal's life in numerous ways. Morphological adaptations often reduce thermal conductance between body and environment. Thick insulation layers, such as fur, reduce heat loss in cold weather conditions, although dense fur can also be used for protection from the sun. Many mammals grow denser and longer fur during the winter to maximize the benefit from insulation. Apart from fur, some mammals use a counter-current heat exchanger to reduce heat loss from their extremities. This is done by a *rete mirabilis*, where highly branched arterioles and venoles come in close contact, heating the blood coming from the extremity and cooling the blood flowing to the extremity. By reducing the temperature gradient between the extremities and the surrounding, less heat is lost via the extremities and more heat is retained in the body core.

Besides morphological adaptations, different behavioural strategies have also evolved. Many mammals build nests and dig burrows. These nests and burrows can be well-buffered against the weather and thus provide a safe sanctuary in adverse weather conditions. Social animals often huddle together with mates in cold conditions. This behaviour reduces the effective body surface of each individual and thus less heat is transferred externally.

Food hoarding is another behavioural adaptation. Although food hoarding has no immediate effect on energy consumption, it liberates the animal from the need to search for food. This is beneficial especially in bad weather and cold temperatures, when the animal can rest in a safe burrow instead of exposing itself to the weather. Furthermore, food availability might be strongly reduced during those times and foraging would take longer, if it was successful at all. Thus food hoarding has a strong effect on ultimate energy consumption (Humphries et al., 2002).

Not only morphological and behavioural adaptations have evolved. The most powerful tool in order to save energy lies within the animal's physiology. Several mammals are capable of temporarily lowering their metabolism in order to conserve energy. A decrease of about 20% in metabolic activity during the resting phase compared to the active phase has been observed in most mammals. This is normally accompanied by a reduction in $T_b$ of 0.5 to 2°C (Heldmaier et al., 2004). Apart from this moderate metabolic reduction during sleep and rest, some mammals can lower their MR drastically, even to levels that are clearly below BMR. A variety of mammals from small shrews to large bears employ these mechanisms.

## 1.4 Hypometabolic states

Hypometabolic states are characterised by a reduction of metabolic activity below resting levels. They are normally accompanied by a reduction in $T_b$ and general lethargy. Depending on the duration of metabolic and $T_b$ reduction, two different states can be distinguished, i.e. daily torpor and hibernation. In daily torpor, the reduction of $T_b$ and MR lasts less than 24 h, while it exceeds 24 h in hibernation (Geiser, 2004). Although daily torpor and hibernation are classified as different states based on their duration, the physiological differences might be more quantitative than qualitative (Wilz and Heldmaier, 2000). Daily torpor and hibernation might share the same evolutionary origin and might represent merely two regions of a physiological gradient of hypometabolism and hypothermia (Lovegrove et al., 2001).

### 1.4.1 General properties of daily torpor and hibernation

Torpor and hibernation are both characterized by alternating normothermic, normometabolic periods and hypothermic, hypometabolic periods. A complete cycle of these can be divided into four distinct phases (Heldmaier, 2003). The first phase is a normothermic, normometabolic period, i.e. a normal activity phase for daily torpor or an arousal for hibernation. This is followed by entry into torpor, during which MR is rapidly reduced and $T_b$ declines. During the subsequent torpor or hibernation bout, MR remains low and $T_b$ follows ambient temperature ($T_a$) closely. The torpor or hibernation bout is ended by the fourth phase, the re-warming phase, during which MR rises rapidly and $T_b$ increases towards normothermic levels. Arousals include both the normothermic and the re-warming phase, i.e. both energetically costly phases.

Periodic arousals are very costly and account for most of the energy used during hibernation (Boyer and Barnes, 1999). These were formerly considered obligatory for hibernators (Carey et al., 2003; Humphries et al., 2003), but Dausmann et al. (2005) showed in the fat-tailed dwarf lemur *Cheirogaleus medius*, that is was not arousals per se, but recurring high $T_b$ which were an obligatory part of hibernation. These lemurs hibernate in the tropics and are exposed to large daily $T_a$ and thus $T_b$ fluctuations. However, if $T_b$ was passively elevated above 30°C regularly, *C. medius* remained in hibernation without arousals for many weeks or even months. Although an arousal or periodically high $T_b$ appears to be essential, its function is still unclear.

French (1985) suggested that at the low $T_b$ normally found in hibernators, some metabolism-linked process must go amiss and that recurring, high $T_b$ is needed to re-establish homeostasis. Different physiological demands have been proposed to explain the need for periodic arousals and high $T_b$. For example, during hibernation, liver and kidney function is diminished or absent, and so metabolic by-products accumulate and have to be eliminated (Deavers and Musacchia, 1980). It might also be impossible for hibernators to sleep at low $T_b$ and an increasing "sleep debt" might force hibernators to warm-up periodically to maintain adequate sleep (Daan et al., 1991; Strijkstra and Daan, 1997; Larkin and Heller, 1996). A third possibility is that periodic high brain temperatures are needed to consolidate memory and preserve neural connections in the central nervous system (Popov and Bocharova, 1992; Popov et al., 1992). Finally, high body temperatures might be needed to temporarily boost the immune system and to counteract bacterial infection because the immune system is depressed during hibernation (Prendergast et al., 2002; Luis and Hudson, 2006).

Physiological regulation of hibernation and daily torpor is also poorly understood. The molecular basis of, and neural signalling in initiating and reversing hibernation are still a mystery (Heldmaier et al., 2004; Carey et al., 2003). Although some progress has been made to elucidate the molecular biology of hibernation, research in this area is still in its infancy (Andrews, 2007).

Energy savings during hibernation and daily torpor are considerable. Metabolic rate during torpor (TMR) can be as little as 1% of RMR (Geiser and Ruf, 1995; Geiser, 2004; Wang, 1978). Even factoring in the costly arousals, daily energy expenditure can be reduced by 90-96% (Heldmaier, 2003). But the physiological basis of this is still under intense debate. The temperature-dependent hypothesis suggests that observed reductions in MR with decreasing $T_b$ are based on Q10-effects of biochemical reactions and that this is why $T_b$ and MR drop concurrently when the animal enters torpor (Snapp and Heller, 1981; Guppy and Withers, 1999; Hammel et al., 1968). Because the former hypothesis can not explain the large MR reductions observed during hibernation, it has been rejected as an exclusive explanation for MR reduction. The temperature-independent hypothesis proposes that MR is actively suppressed or downregulated on entry to torpor and that the fall of $T_b$ is the consequence and not the reason for a reduction of metabolism (Heldmaier and Ruf, 1992; Heldmaier et al., 1993b; Storey, 1997). These two hypotheses are not mutually exclusive and it has been suggested that temperature effects as well as physiological inhibition must be involved in the reduction of MR (Geiser, 1988; Storey and Storey, 1990). Furthermore, MR during hibernation might also be reduced by low thermal conductance of torpid animals (Snyder and Nestler, 1990).

Daily torpor and hibernation can be found in a range of mammals from temperate and arctic (Heldmaier et al., 2004), as well as tropical climates (Kelm and von Helversen, 2007; Dausmann et al., 2004; Lovegrove and Genin, 2008). This indicates that these hypometabolic states are not primarily aimed at cold defence, but instead they seem to be a measure to cope with temporal or seasonal energy and water shortage (Heldmaier et al., 2004). That states of hypometabolism are found in a variety of mammals and are common in monotremes (Grigg et al., 2004, 1989), marsupials (Geiser, 1994; Geiser and Ruf, 1995) and basal placental mammals (Lovegrove et al., 1999a) also indicates that hypometabolism is likely to be a basic property of mammalian physiology (Heldmaier et al., 2004).

### 1.4.2 Hibernation

Hibernation is a physiological and behavioural adaptation to survive seasonal periods of energy shortage by a combination of pre-hibernal energy storage and hibernal metabolic depression (Humphries et al., 2003). Preparations for hibernation must be made during the pre-hibernation period, i.e. adequate energy stores have to be accumulated either by hoarding and storing of food or by building up fat stores in the body. It is thus an obligatory behaviour and part of a fixed seasonal timing program (Heldmaier et al., 2004).

Hibernation bouts always last longer than 24 h and normally increase in length towards the middle of the hibernation season. During the start and end of the hibernation season, hibernation bouts are around one to five days in length. Although hibernators are capable of bouts of less than a day in length (Geiser and Kenagy, 1988; Song et al., 1997; French, 1985), it appears that these bouts are different from the daily torpor bouts they resemble. Functionally they can be considered as short hibernation bouts (Geiser, 1988; Geiser and Brigham, 2000; Wilz and Heldmaier, 2000). Hibernation bouts last up to 20 d in the middle of the hibernation season (Heldmaier, 2003) and extremely long hibernation bouts of up to 45 d have been found in the European badger *Meles meles* (Fowler and Racey, 1988). The length of the hibernation season differs from species to species as well as between habitats (Turbill et al., 2003a; Geiser, 2004; Arnold, 1993). A typical hibernation season might last from a couple of months to over half a year and if held under suitable hibernation conditions in the lab, the pygmy-possum *Cercartetus nanus* can remain in hibernation for over a year (Geiser, 2007).

As mentioned above, metabolic savings during hibernation can be pronounced. Most of the energy required during the hibernation season is consumed during arousals and the euthermic periods. In the alpine marmot *Marmota marmota*, these make up over 70% of the overall energy budget (Heldmaier et al., 1993a). Nevertheless, energy expenditure during hibernation is normally reduced to below 15% of what the animal would have expended if it remained normothermic throughout the winter (Wang, 1978).

### 1.4.3 Torpor

Torpor, or daily torpor, is a controlled reduction of MR and $T_b$ to low levels for periods less than 24 h (Geiser, 2004). It is used by many small mammals in arctic and temperate regions as well as in the tropics. Torpor may be used as a routine energy saving strategy on a daily basis or as an emergency measure when thermoregulatory costs are high during cold periods (Christian and

Geiser, 2007).

Although the animals are normally only torpid during the resting phase, the energy savings achieved are nevertheless considerable. Daily energy savings through the use of torpor are normally about 20-50% because of the high recurring costs of re-warming, even though MR may be reduced by 90% during a torpor bout (Geiser and Ruf, 1995; Lovegrove et al., 1999b). Although energy savings are not as large as during hibernation, daily torpor allows the maintenance of territorial and social activities. Additionally, no pre-hibernal energy reserves have to be accumulated and thus it may be an attractive alternative to hibernation (Heldmaier et al., 2004).

## 1.5 Tropical hibernation

Hibernators are best known for an occur most frequently in arctic and temperate climates, but they are not limited to these regions and also occur in the tropics. Thus far, only three groups of animals are known to hibernate or use daily torpor in the tropics, various bats (Kelm and von Helversen, 2007; Bartels et al., 1998), cheirogaleid lemurs (Dausmann et al., 2004; Schmid, 2001; Kobbe and Dausmann, 2009) and tenrecs (Scholl, 1974; Lovegrove and Genin, 2008).

The main difference between temperate or arctic hibernation and tropical hibernation is the high, and often strongly fluctuating $T_a$ at which hibernation occurs. Avoidance of very low $T_a$, normally found in temperate and arctic regions during winter, is thus not a crucial part of tropical hibernation. The "cold" season in the tropics is usually a season with limited precipitation and resulting limited food and water availability. These seasonal food shortages might be one of the main reasons to use hibernation in the tropics. But as the season or the environment gets drier, the water saving potential of hibernation and daily torpor becomes more significant. Although the primary purpose of daily torpor is energy conservation (Boyer and Barnes, 1999), it may also be important for water conservation, especially in arid-zone species (Macmillen, 1965; Cryan and Wolf, 2003). The reductions in MR, $T_b$, food requirements and activity are accompanied by reductions in evaporative, faecal and urinary water loss, which may lead to substantial water savings (Cooper et al., 2005). Schmid and Speakman (2000) revealed in a field study on the Madagascan grey mouse lemur *Microcebus murinus,* that this species conserves water during the dry season by entering daily torpor.

Arctic and temperate hibernators encounter very low $T_a$ during the hibernation season and subsequently exhibit very low $T_b$. Alpine marmots prefer a hibernation $T_b$ of 5-10°C (Ortmann and Heldmaier, 2000) and other rodents prefer 1-6°C (Heldmaier et al., 2004). The arctic ground squirrel *Spermophilius parryii* can even withstand $T_b$ of -3°C during hibernation (Barnes, 1989). In the tropics on the other hand, $T_a$ is generally above that observed in temperate regions and minimal $T_a$ rarely drops below 5°C (Lovegrove, 2003). Since the reduction in MR seems to be at least in part an effect of lowered $T_b$ (Geiser, 2004), lower $T_a$ during hibernation should lead to larger metabolic reductions. The first example came from a field study on the tropical hibernating primate *C. medius*, that saves around 70% of energy during hibernation compared to the active season (Dausmann et al., 2009). Compared to energy saving of temperate and arctic hibernators, that can conserve up to 90% of energy (Heldmaier, 2003), this is notably lower. If reduced energy savings are a common property of tropical hibernation has to be verified for other tropical hibernators as well. It is thus hypothesized that:

> *Tropical hibernation, i.e. hibernation at high ambient temperatures, is energetically less efficient than hibernation at low ambient temperatures.*

The energetic costs of arousals will be less in warmer conditions, but energy consumption during the hibernation bout will increase. It is thus hypothesized that:

> *At low ambient temperatures tropical hibernators use less energy during hibernation bouts, but require more energy for re-warming during arousals.*

Another characteristic that differs between temperate and tropical hibernation is the course and pattern of $T_a$, or, to be more specific, the temperature within the hibernaculum. Arctic and temperate hibernators usually retreat into caves or deep burrows in the ground. These burrows are well buffered against fluctuations of cold winter temperatures, meaning that arctic and temperate hibernators are exposed to relatively constant (albeit low) $T_a$ (Arnold et al., 1991; Buck and Barnes, 1999). Because $T_b$ resembles $T_a$ of the hibernaculum closely during hibernation, temperate and arctic animals have a relatively constant $T_b$ during hibernation bouts (Arnold, 1993).

Tropical hibernators on the other hand often inhabit poorly insulated hibernacula, either because of the lack of better insulated ones or because of the lack of pressure to have a well insulated hibernaculum (Dausmann et al., 2005). Tree hollows, that are often used as a hibernaculum, have an insulating effect and act as a buffer, but much less pronounced than deep underground burrows (Schmid, 1998). Therefore, tropical hibernators often experience considerable fluctuations of both $T_a$ and $T_b$. Daily $T_b$ fluctuations of up to 20°C have been found in the hibernating lemurs *Microcebus griseorufus* and *C. medius*, these are among the highest found in any hibernating mammal (Kobbe and Dausmann, 2009; Dausmann et al., 2005). These fluctuations may be further exacerbated by heating of the hibernaculum by solar radiation during the day.

Since arousals are very costly and consume most of the energy required over the whole hibernation season (Heldmaier et al., 1993a), any reduction of arousal costs could be beneficial for the animal. Passive heating during arousals, such as using the daily $T_a$ fluctuations, could reduce energy consumed during arousals. The animal would benefit from reduced arousal costs not only during hibernation, but also during daily torpor. That re-warming can be to a large extent be a passive process, using $T_a$ fluctuations, has been shown in several species (Schmid et al., 2000; Lovegrove et al., 1999b; Mzilikazi et al., 2002). Although a strongly fluctuating $T_a$ within the hibernaculum in contrast to a more constant one had no apparent effect on overall MR in the fat-tailed dwarf lemur *C. medius*, arousals were less costly when warming was passively assisted by fluctuating $T_a$ (Dausmann et al., 2009). It is thus hypothesized that:

> *Fluctuating ambient temperatures are energetically beneficial compared with constant ambient temperatures, both for hibernation and daily torpor because of reduced rewarming costs.*

Fluctuating $T_a$ elicit an extraordinary response in *C. medius* (Dausmann et al., 2005). This primate displays a normal hibernation pattern in well insulated tree hollows, where $T_a$ fluctuations are small, and where hibernation bouts are interrupted by periodic arousals. However, if the lemurs choose poorly insulated tree hollows, where $T_a$ fluctuates markedly and exceeds 30°C regularly, they remain torpid for long periods without any arousals. To date *C. medius* is the only mammal showing such a response. Whether other tropical hibernators are capable of a similar behaviour is as yet unknown. On the other hand, daily fluctuations in $T_a$ could lead to more frequent arousals since an arousal might be triggered by rising $T_a$. This has been observed in the mouse lemur *M. murinus*, where torpor is actively terminated when ambient temperature exceeds a threshold temperature (Schmid, 1998). This would not only change the pattern of hibernation, i.e. arousal frequency and hibernation bout length, but also increase the overall energetic costs. It is thus hypothesized that:

> *Arousal frequency and hibernation bout length are affected by fluctuating ambient temperatures.*

To answer the above established hypotheses, the lesser Madagascan hedgehog-tenrec was used as a model organism for tropical hibernation because it is the only tropical hibernator, that hibernates with certainty under laboratory conditions and it is easy to keep in captivity.

## 1.6 The lesser hedgehog tenrec

The lesser Madagascan hedgehog tenrec (*Echinops telfairi*; Martin, 1838) is a small insectivorous mammal endemic to Madagascar. Superficially it resembles a European hedgehog, although with a length of 14-18 cm and an average weight of 140g it is considerably smaller (fig. 1.6.1). Its belly is sparsely haired and its back is armoured with rigid spines, ranging in colour from pale grey to nearly black. If threatened, it can curl into a ball with head and legs tucked inside, only exposing its spiny back.

*E. telfairi* occurs in the south-western parts of Madagascar (fig. 1.6.2), where it inhabits deciduous, gallery and xerophytic spiny forests (Garbutt, 2007). Despite its appearance, *Echinops* is a good climber and leads a semi-arboreal life. It feeds mainly on insects and fruits in the branches of trees and shrubs as well as on the ground (Gould and Eisenberg, 1966). The lesser hedgehog tenrec is a nocturnal or crepuscular animal and rests during the day in tree hollows. Although reputed to be solitary it can also be found in groups of two or three when resting during the day. During its nightly excursions it may be able to orientate itself by echolocation

Figure 1.6.1: The lesser hedgehog tenrec (*Echinops telfairi*)

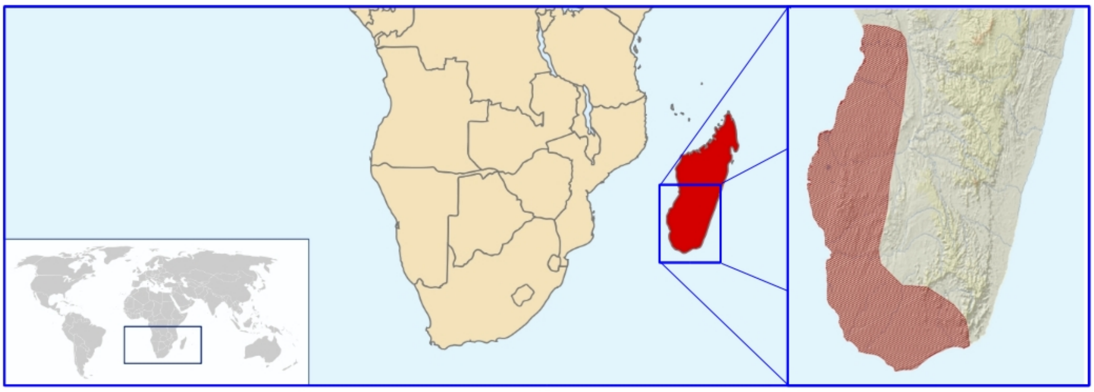

Figure 1.6.2: Distribution of *Echinops telfairi*
The area on Madagascar (red) in which *Echinops telfairi* occurs is shaded red

using tongue clicks (Gould, 1965). Male hedgehog tenrecs are capable of secreting a white liquid used for marking their territory from both nasal and eye-glands (Nicoll, 2001).

The lesser hedgehog tenrec belongs to the family Tenrecidae, which consists of 32 species separated in four subfamilies (see table 1.1 for a full account of the Tenrecidae). The tenrecs had traditionally been placed with shrews, moles, solenodons and golden moles in the insectivoran suborder Soricomorpha (MacPhee and Novacek, 1993), but newer molecular data dispute this classification. Nowadays, the tenrecs and the closely-related golden moles (Chrysochloridae) are placed in the order Afrosoricida (Stanhope et al., 1998a,b). The Afrosoricida do not belong to the Insectivora, but instead are part of the Afrotheria (Nikaido et al., 2003), which is a clade of endemic African mammals that include a variety of diverse mammals, such as elephants, hyraxes, manatees, aardvarks and elephant shrews. *E. telfairi* is thus more closely related to the elephant than to the European hedgehog and is a stunning example of convergent evolution.

The Tenrecidae are a very old mammalian group, that possess basal traits such as a cloaca or internal testicles. The oldest tenrecoid fossils date back to the Miocene (24 million years ago) and were found in sediments from Kenya (Nicoll, 2001). Tenrecs colonized Madagascar after its separation from the African mainland, which occurred 120 to 165 million years ago (Rabinowitz et al., 1983). How they dispersed to Madagascar is unclear. McCall (1997) proposed a land bridge across the Mozambique channel, but newer research indicates that tenrecs and most other Madagascan mammals likely drifted on ocean currents on vegetation rafts to Madagascar (Ali and Huber, 2010). Also unresolved is the question of whether there have been multiple colonization events and whether the Tenrecidae are monophyletic or paraphyletic. Asher (1999) favours a multiple colonization scenario with a paraphyletic origin of the Tenrecidae, whereas the molecular phylogenetic analyses from Douady et al. (2002) provide support for the monophyly of the Tenrecidae and Malagasy tenrecs.

Table 1.1: Systematic classification of the Tenrecidae

Family Tenrecidae - Tenrecs

- Subfamily Tenrecinae - Spiny tenrecs:
  - greater hedgehog tenrec (*Setifer setosus*)
  - lesser hedgehog tenrec (*Echinops telfairi*)
  - common tenrec (*Tenrec ecaudatus*)
  - lowland streaked tenrec (*Hemicentetes semispinosus*)
  - highland streaked tenrec (*Hemicentetes nigriceps*)
- Subfamily Geogalinae - Large-eared tenrecs:
  - large-eared tenrec (*Geogale aurita*)
- Subfamily Oryzorictinae - Furred tenrecs:
  - aquatic tenrec (*Limnogale mergulus*)
  - rice tenrecs (*Oryzorictes*, 2 species)
  - shrew tenrecs (*Microgale*, 20 species)
- Subfamily Potamogalinae - Otter shrews:
  - otter shrews (*Micropotamogale*, 2 species)
  - giant otter shrew (*Potomogale velox*)

*E. telfairi* is considered a proto-endotherm, as it has the lowest normothermic $T_b$[5] and displays the most primitive form of adaptive heterothermy of all placental mammals (Lovegrove and Genin, 2008). It uses daily torpor obligatory in the summer, i.e. it lacks periods of normothermy lasting more than 24 h and enters torpor every day (Scholl, 1974). Nevertheless, it can produce endogenous heat and females can maintain a high normothermic $T_b$ for up to one month during the breeding season (Poppitt et al., 1994). In winter the lesser hedge-hog tenrec hibernates and hibernation bouts last on average 3 to 4 d, with the longest observed hibernation bout lasting

---

[5] an exception to this is the naked-mole rat *Heterocephalus glaber*, that is unable of effective thermoregulation and is considered an "poikilothermic" mammal (Woodley and Buffenstein, 2002)

11 d (Scholl, 1974).

## 1.7 Summary of hypotheses

1. Tropical hibernation, i.e. hibernation at high ambient temperatures, is energetically less efficient than hibernation at low ambient temperatures.
2. At low ambient temperatures tropical hibernators use less energy during hibernation bouts, but require more energy for re-warming during arousals.
3. Fluctuating ambient temperatures are energetically beneficial compared with constant ambient temperatures, both for hibernation and daily torpor because of reduced re-warming costs.
4. Arousal frequency and hibernation bout length are affected by fluctuating ambient temperatures.

# Chapter 2

# Methods

## 2.1 Animals

Twenty-seven lesser Madagascan hedgehog-tenrecs (*Echinops telfairi*) were used for the experiments. The animals were laboratory bred and accustomed to human contact. All animals were earmarked or marked with an injectable micro transponder (ID-100, Trovan ltd., Hessle, UK). Age, sex and mass of the animals are summarized in table 2.1.

Table 2.1: Age, sex and mass of the animals

| | | age (years) | | mass (g) | |
|---|---|---|---|---|---|
| | number | MD | R | ME | R |
| male | 9 | 4 | 4 - 5 | 136 | 118 - 156 |
| female | 18 | 4 | 3 - 5 | 135 | 95 - 195 |
| total | 27 | 4 | 3 - 5 | 135 | 95 - 195 |

MD = median, R = range, ME = mean average

### 2.1.1 Animal care and housing

The animals were kept in an 8 m$^2$ climate chamber (3 x 2.5 m) equipped with day-light lamps[1]. Day-and-night cycle, ambient temperature and relative humidity were accurately regulated (for climate details see table 2.2). The summer and winter period were reversed relative to that prevailing in Madagascar[2] because the animals had been previously kept under these conditions.

---

[1] A preliminary study was conducted in a laboratory during the preceding winter. Ambient temperature and day-and-night cycle were kept according to the values given for the climate chamber. However, relative humidity could not be controlled and the day-and-night cycle was not shifted.

[2] Since the natural habitat of hedgehog tenrec is in the southern hemisphere, it is summer there when it is winter in the northern hemisphere.

Table 2.2: Climate parameters within a seasonal cycle

| | daylight | | temperature | humidity |
|---|---|---|---|---|
| month | (h per day) | from - till | (°C) | (rel. %) |
| January | 11 | 6:30 - 17:30 | 20 | 40 |
| February | 11 | 6:30 - 17:30 | 20 | 50 |
| March | 12 | 6:00 - 18:00 | 22 | 60 |
| April | 13 | 5:30 - 18:30 | 22 | 70 |
| May | 13 | 5:30 - 18:30 | 25 | 70 |
| June | 14 | 5:00 - 19:00 | 25 | 70 |
| July | 13 | 5:30 - 18:30 | 25 | 70 |
| August | 13 | 5:30 - 18:30 | 25 | 60 |
| September | 12 | 6:00 - 18:00 | 22 | 50 |
| October | 11 | 6:30 - 17:30 | 22 | 50 |
| November | 11 | 6:30 - 17:30 | 22 | 40 |
| December | 10 | 7:00 - 17:00 | 20 | 40 |

To limit disturbance of the animals, the day-and-night cycle was shifted by 12 h to allow feeding and handling during the animals' activity phase (twilight and night).

Two or three animals (at most one male per group) were kept together in modified steel rabbit cages (fig. 2.1.1). The cages measured 60 x 45 x 30 cm (l x w x h) and were made of stainless perforated steel plates on which the animals could climb. The steel tray at the bottom of the cage was filled with aspen tree litter or bark mulch. A lockable front door allowed easy access into the cage. Each cage was fitted with a plastic running wheel and wooden frames for climbing. At least one nest box and one additional place of concealment was provided per cage. The nest boxes (14 x 20 x 14 cm) were made of wood and were equipped with a divider in the middle of the box and a hinged lid to allow easy access to animals in the box (fig.2.1.1).

Fresh water was supplied *ad libitum* in a ceramic cup. The animals were fed at least three times each week with a diverse diet consisting of living insects (mealworms, cockroaches and zophobas), wet canned cat food, dry dog food, dry hedgehog food, boiled egg and fresh fruit (banana and grapes).

## 2.2 Experimental procedure

The experiments took place between October 2008 and June 2009 in the biocentre "Klein Flottbek" of Hamburg University, Germany[3]. The control measurements during summer were con-

---

[3] The preliminary study took place during winter 2007/2008 in the biocentre "Grindel" of Hamburg University. Of the 10 animals that were investigated in the condition $W_{const}$, only four were included in the analysis.

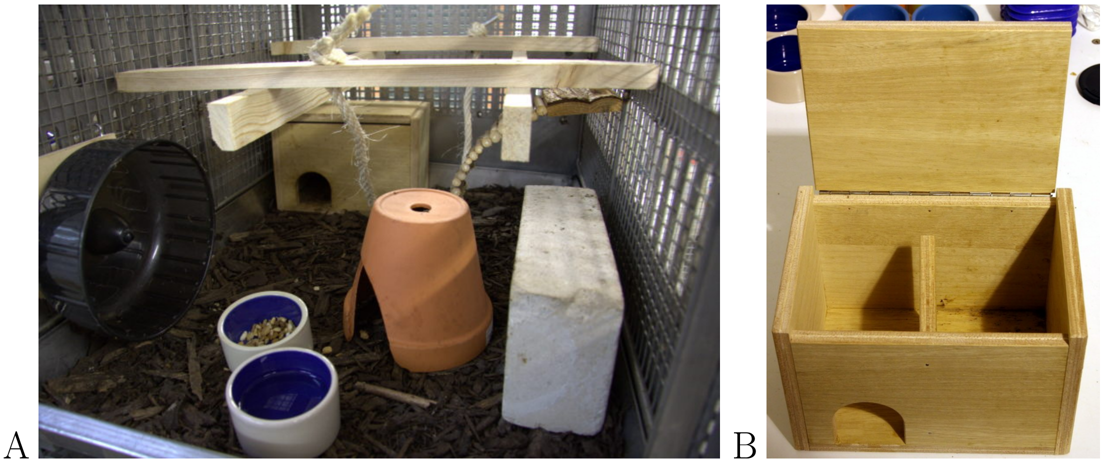

Figure 2.1.1: Furnishing of the cages (A) and a wooden nestbox (B)

ducted in the same climate chambers as those described above. For all other experiments the animals were moved to a smaller climate chamber (1,5 x 2,5 m) equipped similarly to the climate chamber they were housed in. The animals were placed in wooden cages (35 x 21 x 35 cm) equipped with identical nest boxes, tree litter and food and water cups. After the animals had been weighed and equipped with temperature loggers (for details see chapter 2.3.2) they were placed in the nest boxes and the measurements started.

A total of five different experimental conditions were investigated (figure 2.2.1 summarizes the different conditions). Each experimental condition lasted for a minimum of two weeks and the sequence of experimental conditions was randomized. Animals were randomly assigned to the different experimental conditions and were used in multiple experiments. Table 2.3 shows the number of animals (N) and days measured (n) for each condition, separated into oxygen consumption ($VO_2$) and $T_b$ measurements. Due to technical problems with the equipment, faulty and missing data, not all data sets were used for the analyses.

During the summer the animals were investigated at a constant $T_a$ of 25°C ($S_{const}$) and at fluctuating $T_a$ between 20°C at night (11 h) and 30°C during the day (13 h, $S_{fluc}$). During the winter the animals were exposed to constant $T_a$ of 15°C ($W_{low}$) and 20°C ($W_{const}$). A further experimental condition included fluctuating $T_a$ between 15°C at night (13 h) and 25°C during the day (11 h, $W_{fluc}$).

Table 2.3: Number of animals/groups and days measured/analysed for all experimental conditions, divided by $VO_2$ and $T_b$ measurements

| | | data acquisition | | | data analysis | | |
|---|---|---|---|---|---|---|---|
| condition | $T_a$ | N | n ($T_b$) | n ($VO_2$) | N | n ($T_b$) | n ($VO_2$) |
| $W_{const}$ | 20°C | 15 | 307 | 135 | 15 | 172 | 115 |
| $W_{fluc}$ | 15-25°C | 14 | 160 | 160 | 12 | 75 | 71 |
| $W_{low}$ | 15°C | 8 | 102 | 96 | 8 | 93 | 0 |
| $S_{const}$ | 25°C | 21 | 165 | 118 | 10 | 40 | 31 |
| $S_{fluc}$ | 20-30°C | 11 | 228 | 232 | 8 | 33 | 30 |

N = number of animals/groups, n = number of days measured/analysed

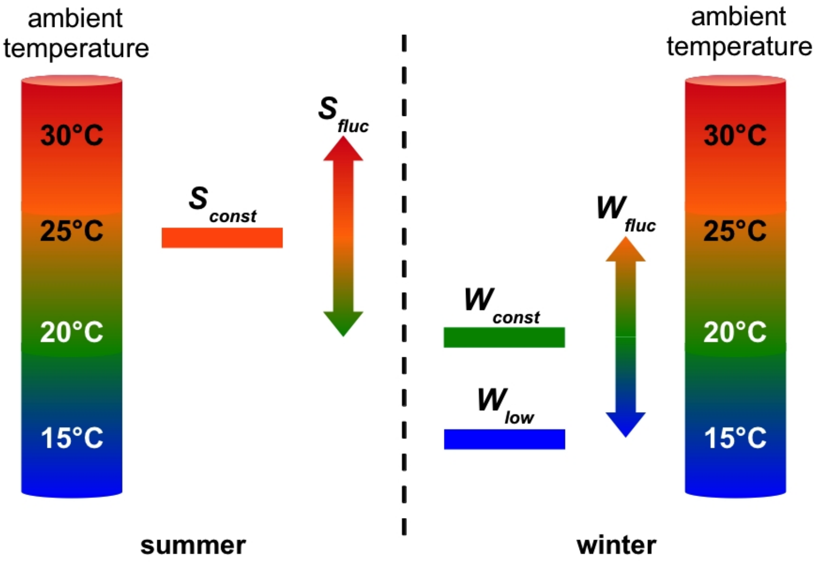

Figure 2.2.1: Summary of experimental conditions

$S_{const}$: 25°C, $S_{fluc}$: 20°C-30°C, $W_{const}$: 20°C, $W_{low}$: 15°C, $W_{fluc}$: 15°C-25°C

## 2.3 Measured parameters

### 2.3.1 Oxygen consumption

#### 2.3.1.1 Construction and setup of the respirometry system

Oxygen consumption was measured with two differential oxygen analyzers (OxBox2 and OxBox3, FIWI, Vienna University, Vienna, Austria). OxBox2 was a two-channel differential oxygen analyzer with one channel analyzing measurement air and the other channel analyzing reference (ambient) air to allow for correction of temperature drift. The updated model Oxbox3 consisted only of one analysis channel. In both instruments, the measuring air channel switched to reference air for five minutes once per hour for null balancing.

Figure 2.3.1 shows a schematic setup of the complete respiratory system including the nest boxes. The open-circuit respirometry systems included a flow pump that was regulated via a feedback loop by the mass flow meter of the oxygen analyzer and this kept the set flow rate constant. The air was drawn at a rate of 50 l/h from the nest boxes which served as respiratory chambers and thus were fitted with adapters to accommodate the tubes of the respirometry system. To lessen air mixing between ambient air and the inside of the nest box, a divider was installed in the middle of the box. A gas splitter (RM Gas Flow Multiplexer, Sable Systems, Las Vegas, USA) was used to rotate air flow amongst four different nest boxes. Thus a total of eight animals could be measured at the same time with two oxygen analyzers. During the one hour long rotation cycle, all nest boxes were measured for 15 min. Once per hour, reference air was measured for five minutes to obtain a baseline value. Depending on the time that the oxygen analyser started and the timing of the gas splitter, the reference air measurement fell into the measurement period of different animals. Before air entered the analyzer, it was dried by passing it through a tube filled with silica gel (Silica Gel Orange, 2-5 mm, Carl Roth, Karlsruhe, Germany). Depending on ambient conditions and activity of the animals, the desiccant was replaced every two to four days. After the air destined for measurement entered the oxygen analyzer, an aliquot (ca. 20 l/h) was drawn for the main airstream and analyzed.

The complete respirometry system was transportable and battery-powered by a standard 12 volt car battery. Gas-tight rubber tubes (Tygon tube ST, Saint-Gobain, Charny, France) were used for all connections between the parts of the respirometry system. Data points were stored once a minute in the internal memory of the analyzer.

#### 2.3.1.2 Calibration

The linear output of the oxygen analyzer was calibrated using fresh air (with an assumed oxygen percentage of 20.96%) and air with reduced oxygen content (19.91% and 18.86%). The latter gas mixtures were produced with a gas-mixing pump (2KM300/a, H. Westhoft GmbH, Bochum, Germany) using fresh air and adding five and ten per cent nitrogen, respectively. From these three points a calibration value corresponding to an oxygen reduction of one per cent was calculated

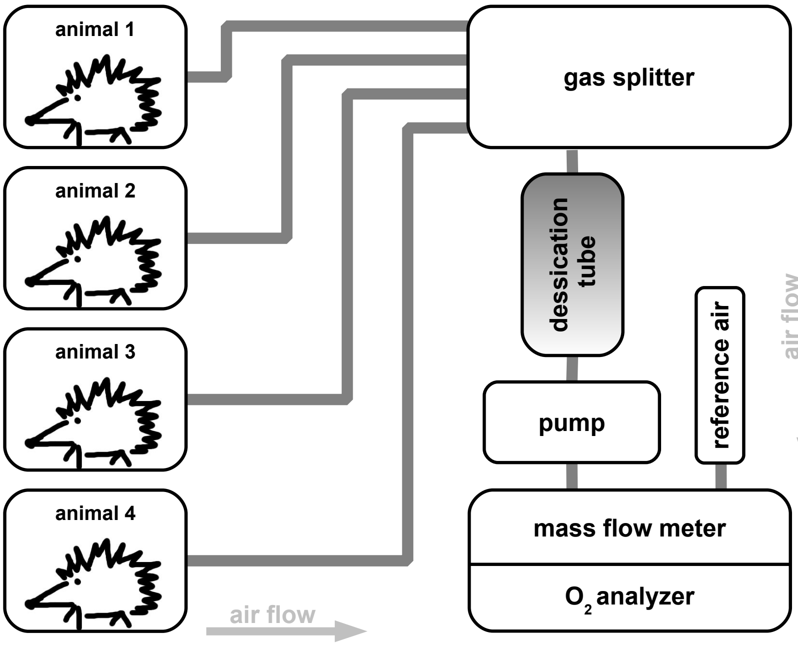

Figure 2.3.1: Schema of the respirometry system

by linear regression and used as the calibration value for analysis.

### 2.3.2 Body temperature

Body temperature was measured with miniaturised temperature data loggers (iButton, DS1922L, Maxim Integrated Products, Inc., Sunnyvale, USA). These were taped to the animals' abdomen with medical tape (Fixomull stretch, BSN medical, Hamburg, Germany). Figure 2.3.2 shows the iButton compared to a 1€ coin and attached to an animal. To improve adherence of the tape, the abdomen was shaved and wiped with alcohol prior to fixation of the tape. Logging interval of the iButtons was set to 15 min. The iButton stayed on the animals' abdomen until it dropped off by itself or was removed after the experiments. If an iButton fell off before the end of the experiment, it was retaped. Attachment of the iButtons was checked by sight every second day. If the iButton was detached at this time, only those data collected up to the point where the iButton was observed attached or at the last arousal/activity phase was included in the analysis.

### 2.3.3 Ambient temperature

Ambient temperature was measured with the same data loggers as $T_b$. In each nest box one iButton was attached to the inside of the lid. An additional temperature logger was fixed to the wooden cage. Logging intervals of $T_b$ and $T_a$ data loggers were synchronized.

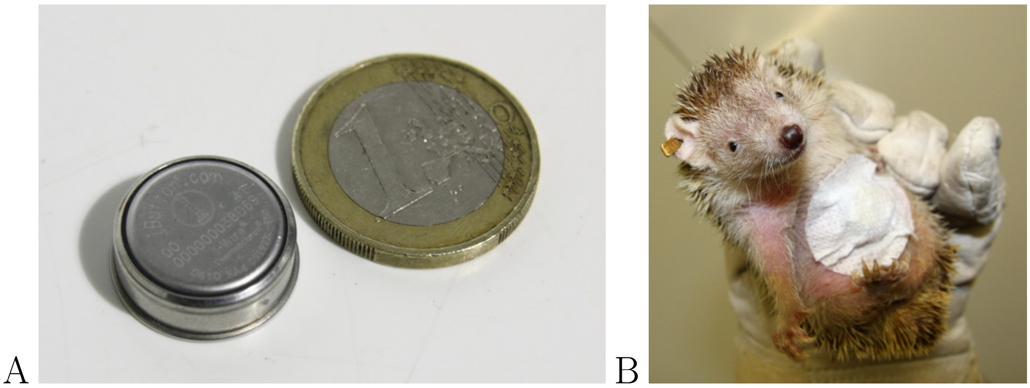

Figure 2.3.2: iButton used for $T_b$-measurements compared to a 1€ coin (A) and attached to a tenrec with medical tape (B)

## 2.4 Data analysis

### 2.4.1 Hibernation bouts, arousals and activity phase

The animals did not show extended periods of normothermy, i.e. $T_b$ fluctuated on a daily basis with high $T_b$ during the activity phase (summer) or arousals (winter) and low $T_b$ during daily torpor bouts (summer/winter) or hibernation bouts (winter). The animals were regarded as normothermic when $T_b$ rose above 28°C.

A daily torpor or hibernation bout was defined as the period when $T_b$ resembled $T_a$ closely, i.e. $T_b$ remained within a limit of two degrees Celsius above $T_a$. When $T_b$ rose above this limit for at least three consecutive hours, the animal was regarded as in an activity phase or arousal (see fig. 2.4.1). The activity phase or arousal started when $T_b$ exceeded the limit for the first time and ended when $T_b$ dropped below the limit again. Start and end time was noted for every daily torpor bout, hibernation bout, activity phase, arousal and normothermic period, and the duration of each was calculated.

### 2.4.2 Temperature

Temperature data were averaged over each hour. Minimum, maximum and average ambient, nest ($T_n$) and body temperature values were calculated for each daily torpor bout, hibernation bout, arousal and activity phase. For arousals and activity phases $T_b$ warming rates were calculated as °C h$^{-1}$. The warming phase was defined as the period from the start of the arousal or activity phase until normothermic $T_b$ ($> 28°C$) was reached.

### 2.4.3 Oxygen consumption

Output values of the oxygen analyzer were corrected for baseline values and drift. The hourly reference air measurements served as baseline values. To account for drift of the oxygen analyzer,

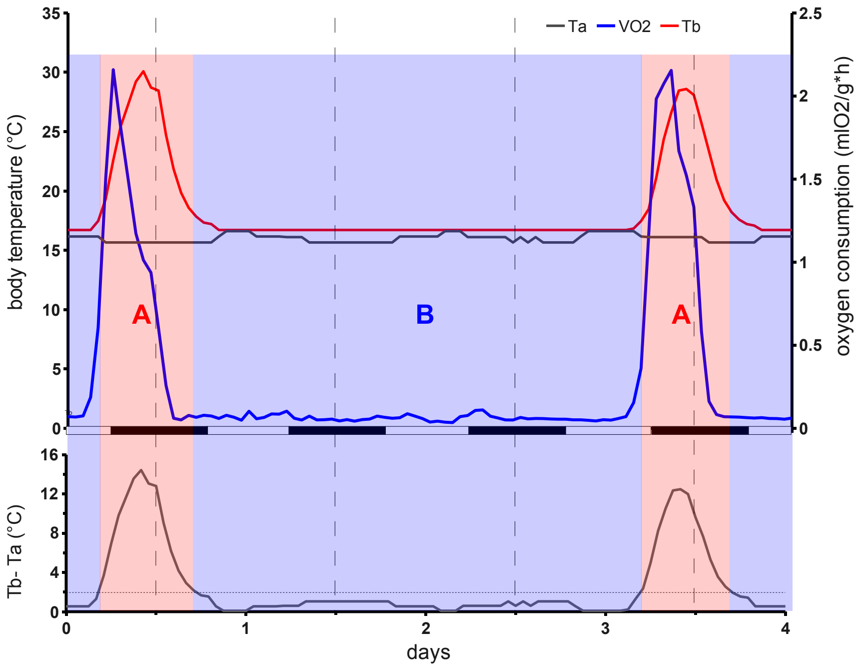

Figure 2.4.1: Definition of hibernation bouts and arousals

Ambient temperature ($T_a$), body temperature ($T_b$) and oxygen consumption ($VO_2$) are shown in the upper part of the diagram, whereas the lower part shows the difference between $T_b$ and $T_a$. An arousal (A) is defined as the time $T_b$-$T_a$ exceeds 2°C (dotted vertical line in the lower part of the diagram) for more than three consecutive hours, otherwise the animal is in an hibernation bout (B). Arousals and hibernation bouts are marked with red and blue boxes, respectively. Vertical dashed lines indicate midnight, whereas white and black areas of the box in the upper x-axis indicate day and night, respectively.

a linear regression between each two consecutive reference air measurements was calculated and the calculated baseline values were subtracted from the corresponding output values.

The difference in oxygen concentration between ambient and measurement air was calculated with the formula:

$$\triangle O_2 = DO_2/Calib$$

**with** $\triangle O_2$: $O_2$ difference between measurement and ambient air in %; $DO_2$: output value of the oxygen analyzer corrected for baseline and drift; Calib: calibration value (see section 2.3.1.2)

and was corrected for the cross-sensitivity of the $O_2$-sensor to exhaled $CO_2$ with formula[4]:

$$\triangle Vol\%O_2 = \triangle O_2 + \triangle O_2 * S_{CO_2}$$

**with** $\triangle Vol\%O_2$: $O_2$ difference between measurement and ambient air in % corrected for $CO_2$ side effect; $S_{CO_2}$: value for the $\triangle O_2$ side effect of 1% $CO_2$ ($S_{CO_2} = 0.0818$)

---

[4] Cross-sensitivity of the $O_2$-sensor ($O_2$-sensor, Bieler + Lang GmbH, Achern, Germany) to $CO_2$ was calculated by estimating the $CO_2$ concentration of the measurement air for an assumed RQ of 0.85 and comparing it to manufacturer information of the $O_2$-sensor (Ruf, 2010, pers. comm.)

Oxygen consumption was calculated as milliliters $O_2$ per hour ($ml_{O_2}h^{-1}$) using the following formula:

$VO_2\left[ml_{O_2}h^{-1}\right] = \triangle Vol\%O_2 * flow\left[lh^{-1}\right] * 10$ (Heldmaier and Ruf, 1992)

and was converted to $VO_2$ per gram body weight ($ml_{O_2}g^{-1}h^{-1}$). Body weight was calculated as the average of body weight before and after the experiments. Oxygen consumption values were averaged over each hour and synchronized with corresponding temperature readings. For each daily torpor bout, hibernation bout, arousal and activity phase minimum, maximum and average oxygen consumption was calculated. Additionally, the average oxygen consumption for the warming phase was calculated. Oxygen consumption reflects resting metabolic rate (RMR) in most cases, but see section 4.1.3 for a full discussion. Daily energy expenditure (DEE) was calculated using a respiratory quotient (RQ) of 0.85, which reflects a metabolic combustion of 50% fat and 50% carbohydrates. At this RQ, the caloric equivalent is 20.37 $kJl_{O_2}^{-1}$. Values for DEE are given in the form of $kJd^{-1}g^{-1}$.

## 2.5 Statistics

All statistical procedures were performed with SPSS 15 (SPSS Inc., Chicago, USA) or R (Fox, 2005; R Core Development Team, 2004). All data were checked for normality with Kolmogorov-Smirnov test and for homogeneity of variance with Levene's test. Comparisons between two experimental conditions were made with Student's t-tests. Comparisons between three or more experimental conditions were made using one-way Analysis of Variance. Pair-wise multiple comparison procedures (Bonferroni method) were employed to identify which variables differed from each other. Significance was accepted at $P < 0.05$.

# Chapter 3

# Results

## 3.1 Summer

Two different conditions were investigated during the summer; the constant $T_a$ condition $S_{const}$ and the fluctuating $T_a$ condition $S_{fluc}$. A summary of all relevant values as well as a statistical summary is given in table 3.3, table 3.4 and table 3.5 at the end of this chapter.

### 3.1.1 Behavior and body mass

The animals were active during the late day and early night throughout summer[1]. They rested inside their nestboxes during daily torpor bouts and were frequently outside the nestboxes during their activity phase. They ate and drank regularly. A separate student study revealed that animals ate 8.2 ± 0.5 g (values are means ± standard deviation; dry weight (DW); N=6, n=18) of mealworms (*Tenebrio molitor*) and 4.1 ± 2.8 g (DW; N=6, n=18) of banana within one month (Lund, 2009). Average body mass during summer was 156 ± 28 g (N=29), ranging from 111 g to 234 g, whereas females (160 g, N=18) were heavier than males (144 g, N=11 ; *t-test*, $t = 2.0739$, $p = 0.047$).

### 3.1.2 Ambient temperature

Preset values for the climatic chambers and recorded ambient temperatures differed. The average recorded $T_a$ was 24.1 ± 1.7 °C for $S_{const}$. For $S_{fluc}$, $T_a$ at night averaged 19.0 ± 0.3 °C and was on average 28.4 ± 0.6 °C during the day. Ambient temperature took two hours to reach night and day values, respectively. The values of this transitional period were excluded for night and day $T_a$ calculations.

---

[1] times refer to the manipulated time, i.e. 12:00 and 24:00 hours are the middle of the day and night phase, respectively

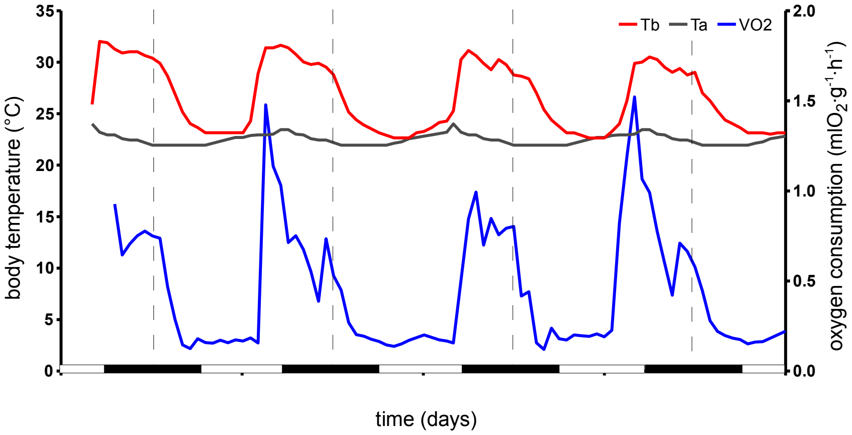

Figure 3.1.1: Daily torpor pattern at constant ambient temperature in summer
Body temperature, $T_a$ and $VO_2$ during daily torpor at constant $T_a$ ($S_{const}$) in animal F0603. Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

### 3.1.3 Daily torpor pattern

In summer, activity phases alternated with daily torpor bouts, i.e. all animals entered torpor every day with one particular exception (see section 3.1.3.1 for details).

#### 3.1.3.1 Constant ambient temperature condition $S_{const}$

A typical sequence of activity phases and daily torpor bouts for $S_{const}$ is shown in figure 3.1.1. At the end of a daily torpor bout (which announced the activity phase), $VO_2$ increased manifold within three or four hours. After reaching its maximum, $VO_2$ remained elevated as long as the animal was normothermic but dropped quickly towards the end of the activity phase. Body temperature followed a similar pattern. With a lag of about one hour relative to $VO_2$, it rose from values close to $T_a$ within two to four hours to a maximum of approximately 30°C. After being elevated for this period, $T_b$ decreased linearly until it approximated $T_a$ at the end of the activity phase. Oxygen consumption remained consistently low and $T_b$ remained close to $T_a$ during the daily torpor bout.

The duration of the daily torpor bouts and activity phases were 11:10 ± 4:00 h (N=10, n=41) and 11:15 ± 8:31 h (N=10, n=31), respectively (fig.3.2.2). One animal (F0604) showed normothermic activity phases exceeding 24 h twice (fig. 3.1.2). These were the only two occasions where this was observed. The two activity phases in that animal lasted 28:00 and 40:00 h. If that animal is excluded from analysis, the duration of the activity phases is reduced to 8:41 ± 2:31 h (N=9, n=36). During the activity phase the animals were normothermic ($T_b$ above 28°C) for 7:50 ± 4:57 h (N=10, n=31). If animal F0604 is excluded from analysis, the duration of normothermy is reduced to 6:22 ± 1:52 h (N=9, n=36).

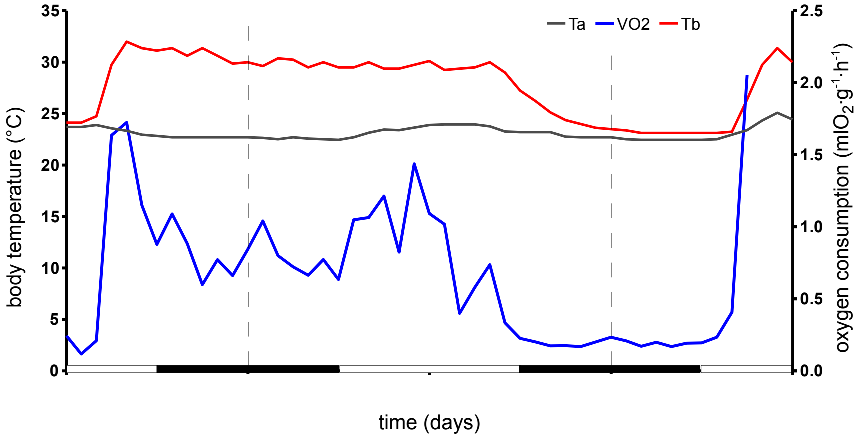

Figure 3.1.2: Activity phase duration exceeding 24 h

Body temperature, $T_a$ and $VO_2$ on one of only two occasions when the activity phase in animal F0604 exceeded 24 h. Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

#### 3.1.3.2 Fluctuating ambient temperature condition $S_{fluc}$

The pattern of daily torpor in fluctuating $T_a$ was similar to that seen in constant $T_a$. An example is shown in figure 3.1.3. During daily torpor bouts, $T_b$ followed fluctuations in $T_a$ passively, but there was a lag between $T_a$ and $T_b$ because of thermal inertia of the animals. Oxygen consumption increased and decreased slightly with rising and falling $T_b$, respectively. During the diurnal phase, $T_b$ followed the increase in $T_a$ passively and $T_b$ reached $T_a$ midway through the diurnal phase. At that point $VO_2$ rose sharply to reach values manifold greater than that during torpor and $T_b$ rose quickly to its maximum and exceeded $T_a$. From then on $T_b$ declined steadily until reaching values close to $T_a$ towards the end of the nocturnal phase.

The duration of the daily torpor bout and the activity phase were 15:16 ± 0:53 h (N=8, n=36) and 6:39 ± 0:36 h (N=8, n=37), respectively (fig. 3.2.2). Duration of daily torpor bouts ($ANOVA$, $F = 29.010$, $p < 0.001$; *Bonferroni*, $p = 1$) and activity phases ($ANOVA$, $F = 1.845$, $p = 0.139$) did not differ between $S_{const}$ and $S_{fluc}$. The animals were normothermic for 4:15 ± 0:26 h (N=8, n=37) during the activity phase. This was shorter compared to $S_{const}$ (*t-test*, $t = 3.282$, $p = 0.009$)[2].

## 3.1.4 Body temperature

### 3.1.4.1 Constant ambient temperature condition $S_{const}$

During daily torpor bouts, $T_b$ averaged 24.6 ± 1.7 °C (N=10, n=41) which was slightly higher than $T_a$ (*t-test*, $t = -4.570$, $p = 0.001$). Mean as well as maximal $T_b$ during the activity phase were always higher than corresponding $T_a$ (*t-test*, $t = -8.263$, $p < 0.001$ for average $T_b$; *t-test*,

---

[2] excluding the two 24+ hour long activity phases of animal F0604 in the $S_{const}$ condition

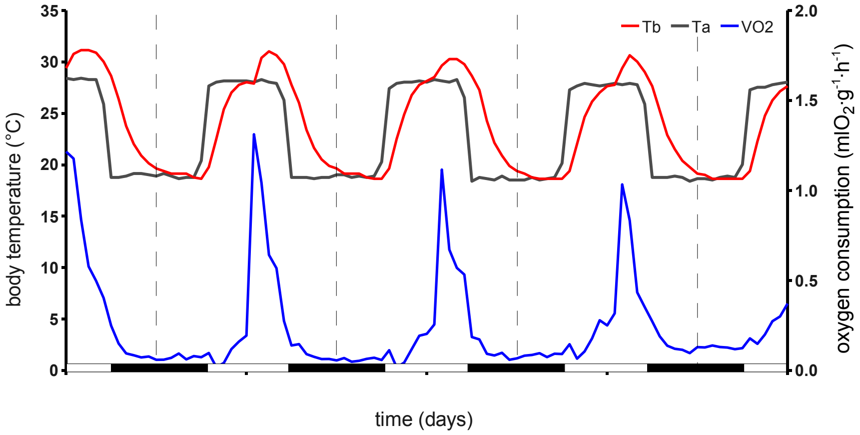

Figure 3.1.3: Pattern of daily torpor in fluctuating ambient temperature in summer

Body temperature, $T_a$ and $VO_2$ during daily torpor at fluctuating $T_a$ ($S_{fluc}$) in animal F0603. Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

$t = -8.873$, $p < 0.001$ for maximal $T_b$). The highest recorded $T_b$ of an animal during the $S_{const}$ condition was 32.8 °C, while mean maximal $T_b$ was 30.8 ± 0.9 °C (N=10, n=31; fig. 3.2.7A). Body temperature over the whole activity phase averaged 28.5 ± 0.8 °C (N=10, n=31) whereas during the normothermic part of the activity phase ($T_b > 28°C$), $T_b$ was 29.8 ± 0.7 °C (N=10, n=31). Maximal $T_b$ and the time when it occurred are shown in figure 3.1.4A. It can be seen from the plot that highest $T_b$ occurred near dusk or during the first half of the night. Warming rate for $S_{const}$ was 2.7 ± 0.5 °C/h (N=10, n=31; fig. 3.2.7B).

#### 3.1.4.2 Fluctuating ambient temperature condition $S_{fluc}$

During fluctuating $T_a$ during the summer, $T_b$ lagged behind $T_a$ because of thermal inertia of the animals. Since $T_a$ changed rapidly between the diurnal and nocturnal phases, the difference between $T_b$ and $T_a$ was most pronounced at the beginning of both day and night.

Maximal $T_b$ during the activity phase averaged 29.8 ± 1.0 °C (N=8, n=37) and the highest observed individual $T_b$ was 31.7 °C. Mean as well as maximal $T_b$ during the activity phase was higher than the corresponding $T_a$ (*t-test*, $t = -6.605$, $p < 0.001$ for average $T_b$; *t-test*, $t = -7.840$, $p < 0.001$ for maximal $T_b$). Average maximal $T_b$ for $S_{fluc}$ did not differ from $S_{const}$ (*ANOVA*, $F = 22.977$, $p < 0.001$; *Bonferroni*, $p = 1$). During the normothermic part of the activity phase, $T_b$ was 29.3 ± 0.7 °C (N=8, n=37) and it averaged 28.4 ± 0.6 °C (N=8, n=37) over the whole activity phase. Compared to $S_{const}$, maximal $T_b$ occurred earlier during the day, at around 14:00 to 16:00 hours, as shown in figure 3.1.4B. The warming rate of 1.5 ± 0.1 °C/h was lower compared to $S_{const}$ (*ANOVA*, $F = 11.516$, $p < 0.001$; *Bonferroni*, $p =< 0.001$).

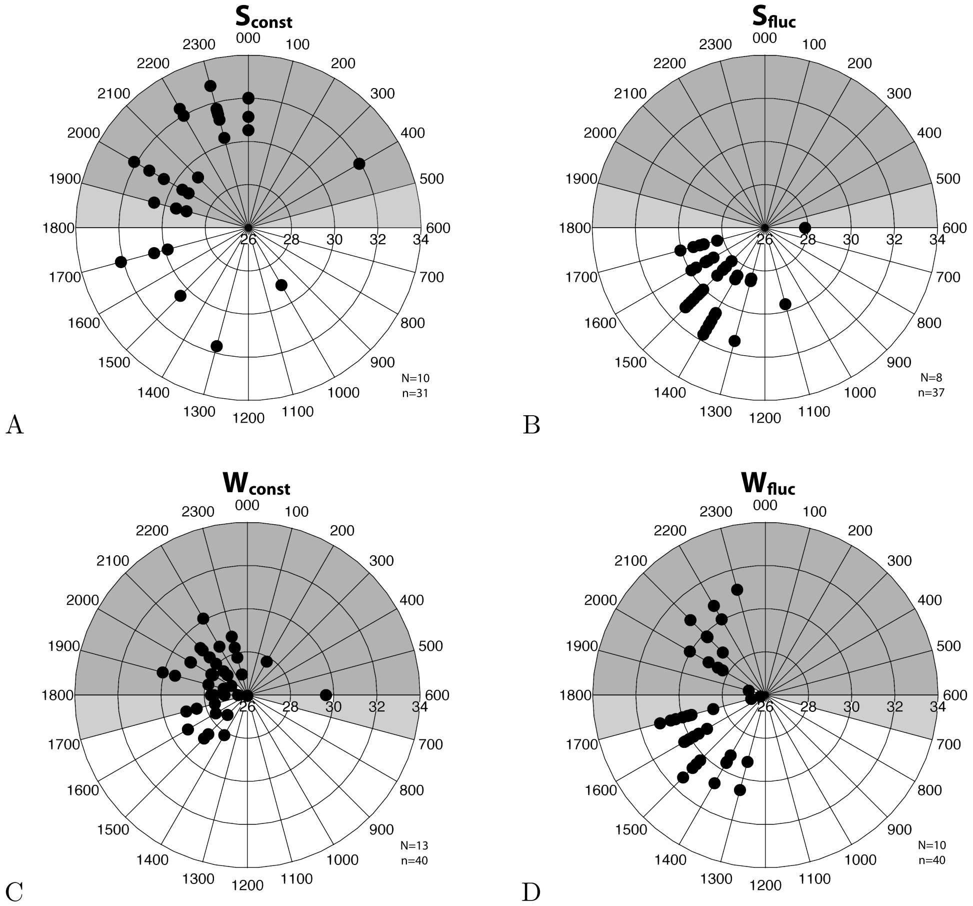

Figure 3.1.4: Circular distribution of maximum body temperature during activity phases and arousals

Time of day (degrees) and level (radial distance) of maximum $T_b$ during activity phases and arousal for $S_{const}$ (A), $S_{fluc}$ (B), $W_{const}$ (C) and $W_{fluc}$ (D). Labels around the circle denote the time (format HHMM) and shaded areas indicate the night phase. Lighter shaded areas are transitional times between night and day phase. Temperature is plotted on the right radius.

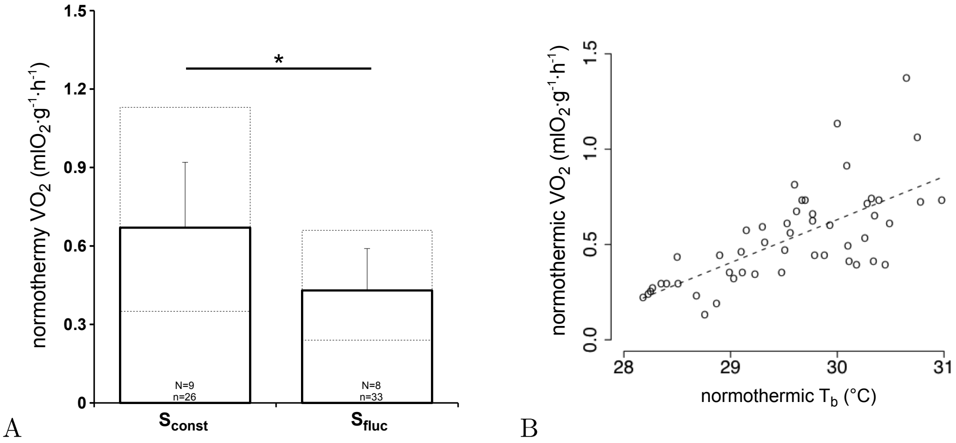

Figure 3.1.5: Oxygen consumption during normothermy and its relation to body temperature
Oxygen consumption during normothermy for $S_{const}$ and $S_{fluc}$(A) and its relation to body temperature (B). Values are means, whiskers indicate the standard deviation and dashed boxes indicate the range in part A, dashed line in part B is the linear regression line (*** = p<0.001, ** = p<0.01, * = p<0.05)

## 3.1.5 Oxygen consumption

### 3.1.5.1 Normothermy

Oxygen consumption of normothermic animals, i.e. $T_b > 28$ °C, was calculated from both the constant $T_a$ $S_{const}$ and fluctuating $T_a$ $S_{fluc}$ experimental conditions. Overall normothermic $VO_2$ is shown in fig. 3.1.5A and averaged 0.56 ± 0.24 $ml_{O_2}g^{-1}h^{-1}$ (N=16, n=59), whereas separate values of $S_{const}$ (0.67 ± 0.25 $ml_{O_2}g^{-1}h^{-1}$; N=9, n=26) were higher than those of $S_{fluc}$ (0.43 ± 0.16 $ml_{O_2}g^{-1}h^{-1}$; N=8, n=33; *t-test*, $t = -2.7948$, $p = 0.027$). As can be seen in fig. 3.1.5B, $VO_2$ during normothermy was positively correlated with $T_b$ (*linear regression*, $VO_2 = 0.226{*}T_b - 6.148$, $R^2 = 0.481$, $F = 44.4$, $p < 0.001$).

### 3.1.5.2 Oxygen consumption during constant ambient temperatures ($S_{const}$)

When the complete activity phase is considered, $VO_2$ averaged 0.55 ± 0.19 $ml_{O_2}g^{-1}h^{-1}$ (N=9, n=26). Maximal $VO_2$ during the activity phase was 1.24 ± 0.55 $ml_{O_2}g^{-1}h^{-1}$ (N=9, n=26), whereas the highest observed $VO_2$ was 3.43 $ml_{O_2}g^{-1}h^{-1}$. The animals consumed 0.73 ± 0.44 $ml_{O_2}g^{-1}h^{-1}$ (N=9, n=26) during the warming phase of the activity phase, i.e. from the start of warming until the animals became normothermic. Oxygen consumption was greatly reduced when an animal commenced daily torpor bouts (*t-test*, $t = 6.670$, $p < 0.001$). Average $VO_2$ during daily torpor bouts was 0.18 ± 0.05 $ml_{O_2}g^{-1}h^{-1}$ (N=9, n=34) which was only one third of that observed during the activity phase. Minimum values of a complete daily torpor bout were 0.12 ± 0.05 $ml_{O_2}g^{-1}h^{-1}$ (N=9, n=34) and the lowest observed value for a complete daily torpor bout was below 0.05 $ml_{O_2}g^{-1}h^{-1}$. The animals consumed on average 0.38 ± 0.21 $ml_{O_2}g^{-1}h^{-1}$ per day (N=9) over the whole length of $S_{const}$, including activity phases and daily torpor bouts.

Table 3.1: Oxygen consumption and percentage of energy saving compared to normothermy for ambient temperature conditions $S_{const}$ and $S_{fluc}$

| | $S_{const}$ | | | $S_{fluc}$ | | |
|---|---|---|---|---|---|---|
| | mean ± SD | energy saving | | mean ± SD | energy saving | |
| normothermy | 0.67 ± 0.25 | - | - | 0.43 ± 0.16 | - | - |
| daily average | 0.38 ± 0.21 | 43 % | * | 0.16 ± 0.05 | 63 % | *** |
| activity phase | 0.55 ± 0.19 | 18 % | n.s. | 0.36 ± 0.12 | 16 % | n.s. |
| torpor bout | 0.18 ± 0.05 | 73 % | *** | 0.08 ± 0.04 | 81 % | *** |

Oxygen consumption is given in $ml_{O_2}g^{-1}h^{-1}$ (*** = p<0.001, ** = p<0.01, * = p<0.05)

This amounted to a DEE of 0.188 ± 0.104 $kJd^{-1}g^{-1}$ (N=9). Table 3.1 summarizes the above values and describes energy savings compared to animals in normothermy (*ANOVA*, $F = 11.246$, $p < 0.001$; *Bonferroni* see table). A graphical display of $S_{const}$ $VO_2$ values is also given in figure 3.2.8.

In figure 3.1.6A $VO_2$ is plotted as a function of $T_b$ over a 24 h period. A consistent pattern was found in most animals. Oxygen consumption rose rapidly during the warming phase (red line) and maximal $T_b$ was reached quickly in around three to four hours. During the cooling phase (black line), $VO_2$ declined steadily with decreasing $T_b$.

#### 3.1.5.3 Oxygen consumption during fluctuating ambient temperatures ($S_{fluc}$)

For $S_{fluc}$, $VO_2$ averaged 0.36 ± 0.12 $ml_{O_2}g^{-1}h^{-1}$ (N=8, n=33) over the activity phase, which was similar to that observed during $S_{const}$ (*ANOVA*, $F = 16.096$, $p < 0.001$; *Bonferroni*, $p = 0.056$). The highest observed maximal $VO_2$ was 1.69 $ml_{O_2}g^{-1}h^{-1}$, while the average maximal $VO_2$ was 0.95 ± 0.28 $ml_{O_2}g^{-1}h^{-1}$ (N=8, n=33), which was the same as for $S_{const}$ (*ANOVA*, $F = 3.490$, $p = 0.029$; *Bonferroni*, $p = 1$). Oxygen consumption during the warming phase was 0.20 ± 0.09 $ml_{O_2}g^{-1}h^{-1}$ (N=8, n=33) which was lower than that observed during $S_{const}$ (*ANOVA*, $F = 19.003$, $p < 0.001$; *Bonferroni*, $p = 0.002$). Oxygen consumption fell to 0.08 ± 0.04 $ml_{O_2}g^{-1}h^{-1}$ (N=8, n=32) during daily torpor bouts. This was only one quarter of the values observed during the activity phase (*t-test*, $t = 7.447$, $p < 0.001$) and was lower than during daily torpor bouts under constant $T_a$ (*ANOVA*, $F = 11.396$, $p < 0.001$; *Bonferroni*, $p < 0.001$). When $VO_2$ was split between the nocturnal and diurnal part of the daily torpor bout, the nocturnal values (0.06 ± 0.04 $ml_{O_2}g^{-1}h^{-1}$; N=8, n=32) were lower than the diurnal values (0.10 ± 0.05 $ml_{O_2}g^{-1}h^{-1}$; N=8, n=32; *t-test*, $t = 5.843$, $p < 0.001$). This difference is also shown in figure 3.2.10. Minimum values of a complete daily torpor bout were 0.03 ± 0.03 $ml_{O_2}g^{-1}h^{-1}$ (N=8, n=32) and the lowest observed value for a complete daily torpor bout was below 0.02 $ml_{O_2}g^{-1}h^{-1}$. Daily $VO_2$ averaged 0.16 ± 0.05 $ml_{O_2}g^{-1}h^{-1}$ (N=8) which corresponds to a DEE of 0.080 ± 0.025 $kJd^{-1}g^{-1}$ (N=8) and was less than that observed during $S_{const}$ (*ANOVA*, $F = 8.099$, $p = 0.001$; *Bonferroni*, $p = 0.006$). Energy savings compared to conditions of

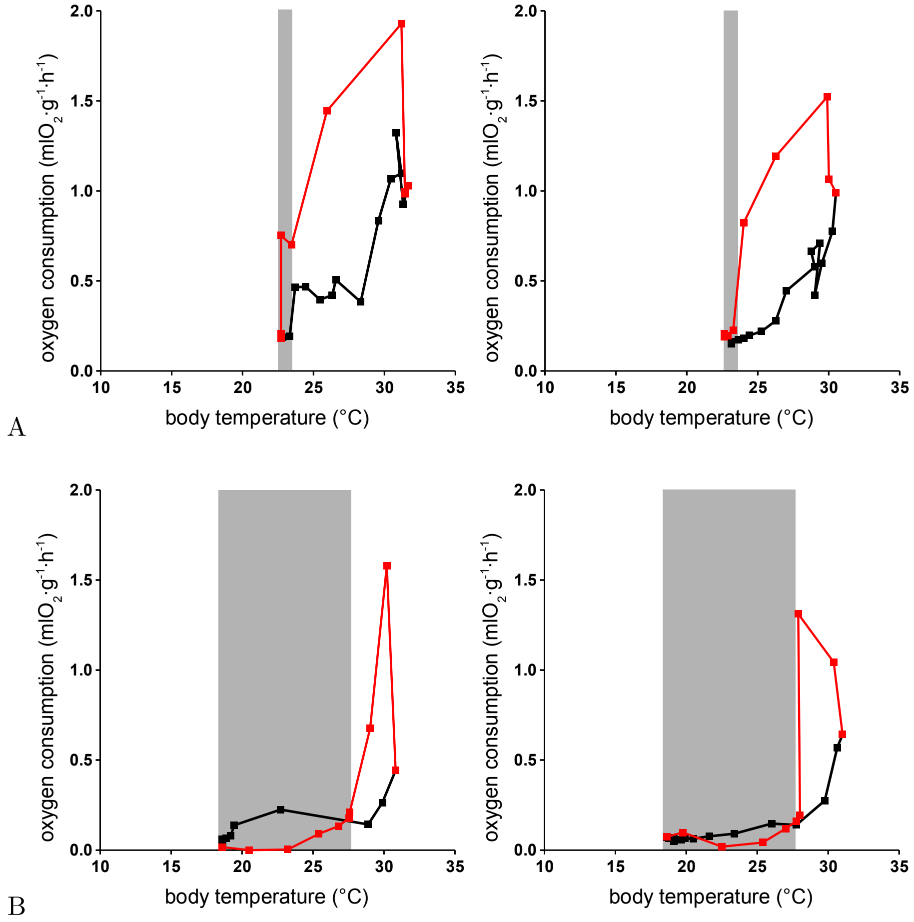

Figure 3.1.6: Oxygen consumption as a function of body temperature during the summer conditions $S_{const}$ and $S_{fluc}$

Oxygen consumption over a 24 h period, including an activity phase and a daily hibernation bout, plotted against corresponding $T_b$ for constant $T_a$ condition $S_{const}$ (A, left: animal F0504, right: animal M0512) and fluctuating $T_a$ condition $S_{fluc}$ (B; left: animal F0408, right: animal: F0508). Red lines and squares are active and passive warming (start to $T_b$ maximum), black lines and squares are cooling ($T_b$ maximum to end). Shaded areas indicate the $T_a$ range over the displayed 24 h period.

normothermy are shown in table 3.1 (*ANOVA*, $F = 20.076$, $p < 0.001$; *Bonferroni* see table). Data on $VO_2$ under other conditions are summarized in figure 3.2.8.

The pattern of $VO_2$ plotted against $T_b$ over a 24 h period is shown in fig. 3.1.6B. The animals warmed up passively using the high $T_a$ during the day, thus $VO_2$ stayed low even with rising $T_b$ (first part of red line). At the point where $T_b$ reached $T_a$, the animals started warming up actively and $VO_2$ rose sharply (second part of red line; see fig. 3.1.3 for the daily pattern). Therefore, the energetic cost for heating was greatly reduced (compare to fig. 3.1.6A). The $VO_2$ and $T_b$ pattern during cooling was similar to $S_{const}$.

## 3.2 Winter

Three different conditions were investigated during the winter, two constant $T_a$ conditions $W_{low}$ and $W_{const}$ and a fluctuating $T_a$ condition $W_{fluc}$. A summary of all relevant values as well as a statistical summary is given in table 3.3, table 3.4 and table 3.5 at the end of this chapter.

### 3.2.1 Behavior and body mass

The animals were lethargic during the winter and spend most time inside their nest boxes. They slept curled into a ball. Normally the animals stayed within their nestboxes even during arousals, but some briefly left their boxes to drink. Most animals did not eat during the winter. If animals ate during the winter it was only a small fraction of the amount they ate during the summer. The monthly average dry food intake was 0.7 ± 0.5 g (N=6, n=18) for mealworms and 0.6 ± 0.6 g (N=6, n=11) for banana (Lund, 2009). In both cases this was less than was eaten in summer (*t-test*, $t = -26.393$, $p < 0.001$ for mealworms; *t-test*, $t = -2.696$, $p = 0.043$ for banana). Mean body mass fell to 126 ± 24 g (N=23) during the winter (*t-test*, $t = 10.616$, $p < 0.001$), with a range of 80 g to 179 g. There was no difference in body mass between males (123 g, N=9) and females (127 g, N=14) during winter (*t-test*, $t = 0.460$, $p = 0.650$).

### 3.2.2 Ambient temperature

Preset values for the climatic chambers and recorded ambient temperatures differed. The average recorded $T_a$ was 12.3 ± 0.1 °C for $W_{low}$ and 17.7 ± 0.6 °C for $W_{const}$. For $W_{fluc}$, average night $T_a$ was 14.1 ± 0.7 °C and average day $T_a$ was 24.3 ± 0.3 °C. Ambient temperature took two hours to reach night and day values, respectively. The values of this transitional period were excluded for night and day $T_a$ calculations.

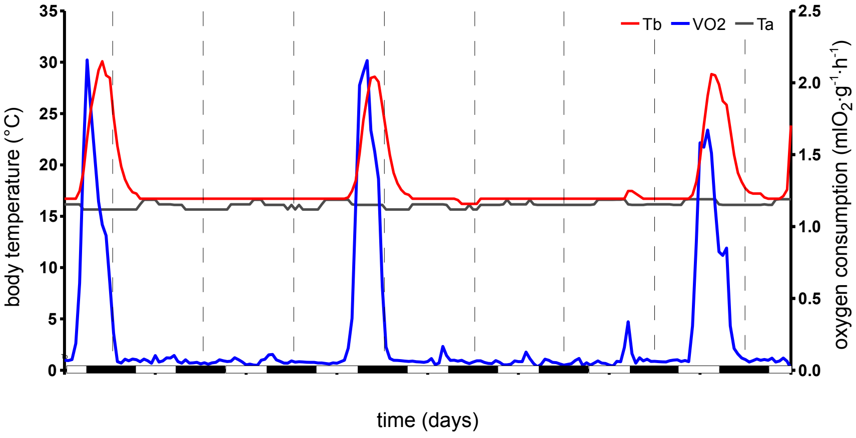

Figure 3.2.1: General hibernation bout and arousal pattern for constant $T_a$

Body temperature, $T_a$ and $VO_2$ during hibernation at constant ambient temperature ($W_{const}$) in animal F0501. Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

### 3.2.3 Hibernation bouts and arousal patterns

#### 3.2.3.1 Constant ambient temperature conditions $W_{const}$ and $W_{low}$

During $W_{const}$ and $W_{low}$ conditions the general pattern of hibernation was similar. Hibernation or torpor bouts of varying lengths alternated with periodic arousals. Fig. 3.2.1 depicts a typical hibernation bout and arousal sequence during constant $T_a$ ($W_{const}$).

At the end of a hibernation bout, prior to arousal, $VO_2$ increased rapidly within three or four hours to values manifold higher than that seen in hibernation. Shortly after reaching maximal values, $VO_2$ declined again and reached values typical of hibernation at the end of the arousal. With a lag of about one hour, $T_b$ rose rapidly over four to six hours from near $T_a$ to approximately 28 °C. After a short period (less than 3 h) in which $T_b$ was maintained at high levels, $T_b$ decreased linearly until it reached $T_a$ at the end of the arousal. During hibernation bouts, $T_b$ remained at ambient levels and $VO_2$ stayed consistently low.

The average duration of hibernation bouts for $W_{const}$ was 4.3 ± 2.2 days (N=16, n=45), with individual bouts ranging from 0.7 to 8.1 d, which was higher than for $S_{const}$ (*ANOVA*, $F = 29.010$, $p < 0.001$; *Bonferroni*, $p < 0.001$). During $W_{low}$, the duration of hibernation bouts increased to 8.5 ± 3.1 d (N=8, n=12*; ANOVA*, $F = 29.010$, $p < 0.001$; *Bonferroni*, $p < 0.001$), with individual bouts ranging from 3.5 to 12.0 d (see fig. 3.2.2A for details). Some hibernation bouts were interrupted by the end of the experiment and therefore could have been longer. If these hibernation bouts are excluded from the analysis, the duration of hibernation bouts changed to 9.2 ± 2.6 d for $W_{low}$ (N=5, n=8). Figure 3.2.3 shows the duration of different hibernation bouts and patterns of hibernation of one animal for all three constant $T_a$ conditions. The duration of hibernation bouts changed dramatically with changing $T_a$, ranging from daily torpor bouts in $S_{const}$ to short hibernation bouts in $W_{const}$ to longer hibernation bouts in $W_{low}$.

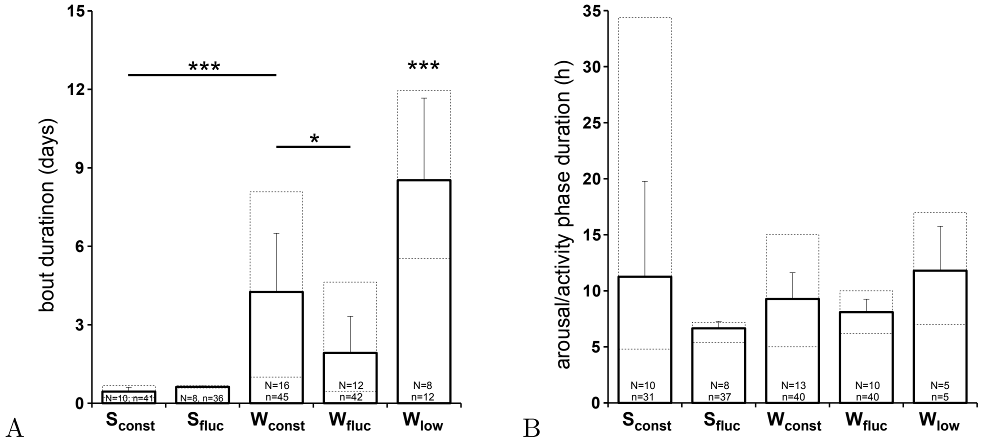

Figure 3.2.2: Hibernation/daily torpor bout and arousal/activity phase duration

Duration of hibernation and daily torpor bouts in days (A) and duration of arousals and activity phases in hours (B) for $W_{low}$, $W_{fluc}$, $W_{const}$, $S_{fluc}$ and $S_{const}$. Values are means, whiskers indicate the standard deviation and dashed boxes indicate the range (*** = $p<0.001$, ** = $p<0.01$, * = $p<0.05$)

The average duration of arousals was 9:16 ± 2:21 h (N=13, n=40) for $W_{const}$ and 11:48 ± 3:57 h (N=5, n=5) for $W_{low}$, respectively (fig. 3.2.2B). No differences in the duration of arousals and activity phases were found between the different conditions (*ANOVA*, $F = 1.845$, $p = 0.139$).

### 3.2.3.2 Fluctuating ambient temperature condition $W_{fluc}$

The general pattern of hibernation bouts and arousals during the fluctuating $T_a$ condition $W_{fluc}$ is shown in figure 3.2.4A.

During hibernation bouts, $T_b$ followed fluctuating $T_a$ with a lag due to thermal inertia of the animals. The animals warmed and cooled passively, whereas $T_b$ reached the low $T_a$ towards the end of the nocturnal phase, but remained slightly below the high $T_a$ at the end of the diurnal phase. Oxygen consumption during hibernation was low during the nocturnal phase and rose with rising $T_b$ during the day, but remained below arousal values.

An arousal was characterized by a sudden increase of $VO_2$ during the middle of the day, when $T_b$ was already elevated and would have almost reached the maximum via passive heating. Following the increase of $VO_2$, $T_b$ rose quickly to values distinctly above $T_a$. After peaking at about 29°C, $T_b$ declined linearly until reaching low $T_a$ at the end of the nocturnal phase.

The average duration of hibernation bouts for $W_{fluc}$ was 1.9 ± 1.4 d (N=12, n=42), with the duration of an individual bout ranging from 0.5 to 4.6 d (fig. 3.2.2A). This was shorter compared that observed in $W_{const}$ conditions (*ANOVA*, $F = 29.010$, $p < 0.001$; *Bonferroni*, $p = 0.017$) but did not differ from $S_{fluc}$ conditions (*ANOVA*, $F = 29.010$, $p < 0.001$; *Bonferroni*, $p = 1.000$). The duration of arousals averaged 8:05 ± 1:09 h (N=10, n=40; fig. 3.2.2B).

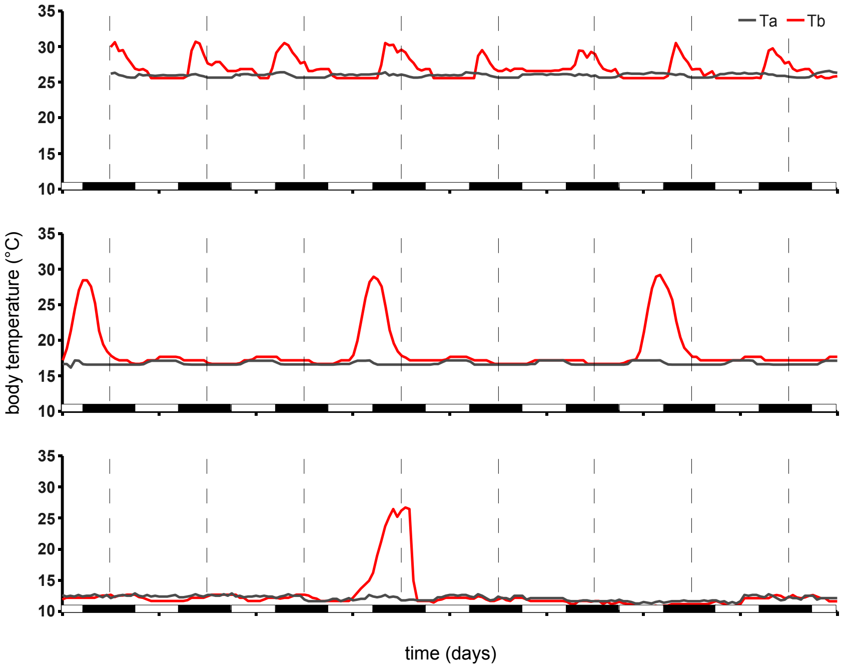

Figure 3.2.3: Hibernation/ daily torpor pattern in constant ambient temperature conditions

Body temperature and $T_a$ of animal M0506 for eight days during $S_{const}$ (top panel), $W_{const}$ (middle panel) and $W_{low}$ (bottom panel). Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

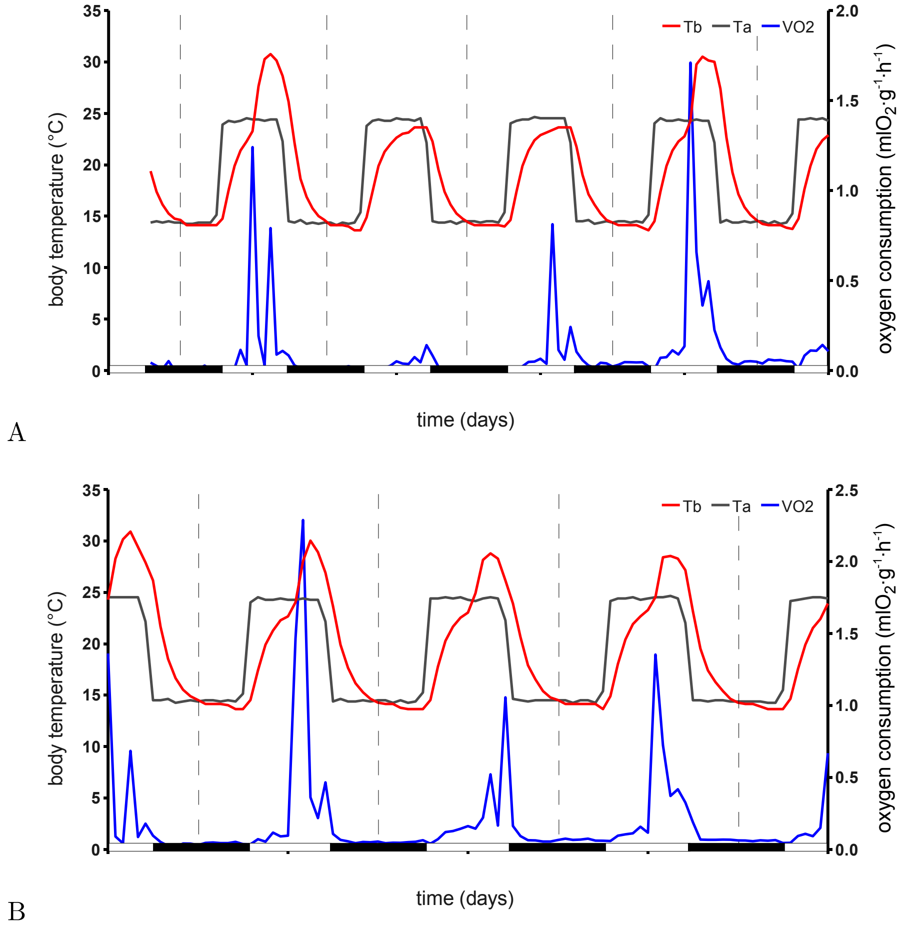

Figure 3.2.4: Hibernation bout and arousal pattern for fluctuating $T_a$ condition $W_{fluc}$

Body temperature, $T_a$ and $VO_2$ during hibernation at fluctuating $T_a$ ($W_{fluc}$) in animal F0508 (A) and M0510 (B). Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

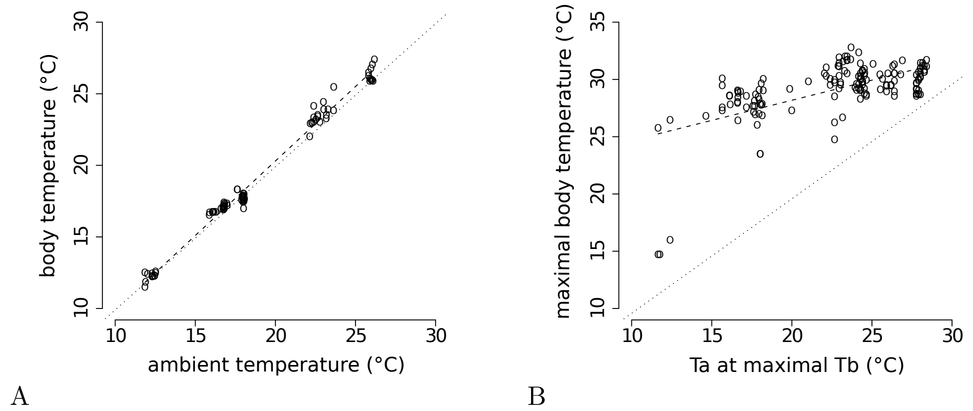

Figure 3.2.5: Body temperature during hibernation and daily torpor bouts versus ambient temperature (A) and maximal body temperature at arousal versus ambient temperature (B) Hibernation and daily torpor bout $T_b$ versus $T_a$ for $W_{low}$, $W_{const}$ and $S_{const}$(A) and maximum arousal $T_b$ versus $T_a$ for all conditions(B). The dotted line represents the bisectrix and the slashed lines represent the linear regression line.

### 3.2.4 Body temperature

#### 3.2.4.1 Constant ambient temperature conditions $W_{const}$ and $W_{low}$

Body temperature followed ambient temperature closely during hibernation bouts. When plotted against each other (including the values of $W_{low}$, $W_{const}$ and $S_{const}$), the resulting linear regression line had a slope close to one ($T_b = 1.040 * Ta - 0.514$, $R^2 = 0.988$, $F = 6685$, $p < 0.001$). Figure 3.2.5A shows this relationship. Consequently, mean $T_b$ did not differ from mean $T_a$ and was 12.7 ± 0.2 °C (N=8, n=12) and 17.6 ± 0.4 °C (N=16, n=45) for $W_{low}$ and $W_{const}$, respectively (*t-test*, $t = -0.100$, $p = 0.923$ for $W_{low}$; *t-test*, $t = 1.227$, $p = 0.239$ for $W_{const}$).

The average $T_b$ at arousal was always higher than the corresponding $T_a$ (*t-test*, $t = -3.072$, $p = 0.037$ for $W_{low}$; *t-test*, $t = 22.863$, $p < 0.001$ for $W_{const}$). In figure 3.2.5B maximum $T_b$ is plotted against average $T_a$ (including the values of all conditions). A linear regression yielded $T_b = 0.351*Ta+21.157$ ($R^2 = 0.367$, $F = 88.07$, $p < 0.001$). When the three outlying points (see below for details) were omitted from analysis, the linear regression yielded $T_b = 0.221*Ta+24.349$ ($R^2 = 0.323$, $F = 70.95$, $p < 0.001$). These three outlying points (belonging to the $W_{low}$ condition) were thought to be “unsuccessful” arousals, based on the criteria for arousal described in the methods section. But instead of reaching a usual maximal $T_b$ of over 25 °C, the animals' $T_b$ remained with 15 °C only sightly above $T_a$. Figure 3.2.6 shows an “unsuccessful” arousal of animal F0501 (A) compared to a successful arousal of animal M0507 (B). During this event $T_b$ of animal F0501 reached a maximum of only 14.7 °C, which was 3.1 °C above $T_a$. The arousal lasted for nine hours (indicated by the shaded red box), during which the difference between $T_b$ and $T_a$ was always more than 2 °C. The successful arousal (B) was uncharacteristic as well, as

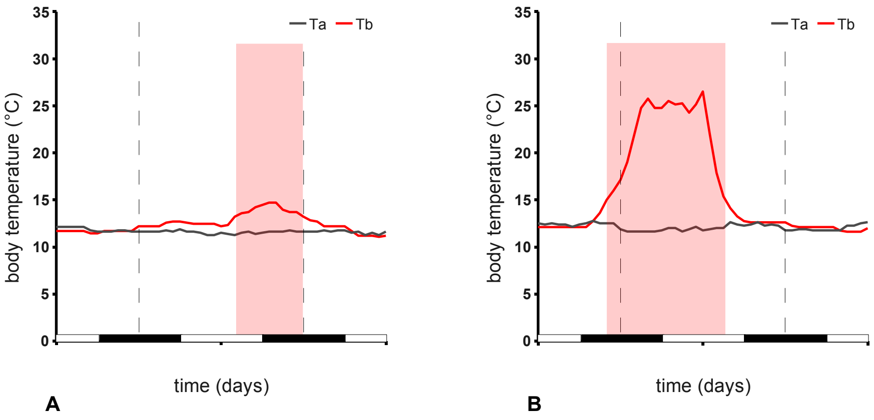

Figure 3.2.6: "Unsuccessful" and successful arousals

Body temperature and ambient temperature of an example for an "unsuccessful" arousal of animal F0501 at Ta15 (A) compared to a successful arousal of animal M0507 at Ta15 (B). The red shaded boxes indicate the arousals, i.e. time when Tb exceeded Ta by more than 2°C for at least three consecutive hours. Vertical dashed lines indicate midnight, whereas white and black boxes on the x-axis indicate day and night, respectively.

it lasted for 17 h, which is 5 h above average for $W_{low}$ (see fig. 3.2.3 bottom panel for another $W_{low}$ arousal). Furthermore, it was the longest arousal observed under all conditions, apart from the two arousals exceeding 24 h described in section 3.1.3.1.

Average maximal arousal $T_b$ was 19.6 ± 6.3 °C (N=5, n=5) and 28.1 ± 1.4 °C (N=13, n=40) for $W_{low}$ and $W_{const}$, respectively (fig. 3.2.7A). When the two "unsuccessful" arousals were excluded, maximal $T_b$ for $W_{low}$ increased to 23.1 ± 6.1 °C (N=3, n=3). Maximal $T_b$ of $W_{low}$ was lower than maximal $T_b$ of $W_{const}$ (*ANOVA*, $F = 22.977$, $p < 0.001$; *Bonferroni*, $p < 0.001$). No difference in maximal $T_b$ was found between $W_{const}$ and $S_{const}$ (*ANOVA*, $F = 22.977$, $p < 0.001$; *Bonferroni*, $p = 0.063$). The highest observed $T_b$ during an arousal in any individual was 26.7°C for $W_{low}$ and 30.1°C for $W_{const}$. Maximal arousal $T_b$ for $W_{const}$ occurred two to three hours before the start of the night or during the first half of the night (see fig. 3.1.4C). Warming rate was 2.7 ± 0.6 °C/h for $W_{const}$ and did not differ compared to that observed in $S_{const}$ (*ANOVA*, $F = 11.516$, $p < 0.001$; *Bonferroni*, $p = 1$; fig. 3.2.7B).

#### 3.2.4.2 Fluctuating ambient temperature condition $W_{fluc}$

Body temperature followed $T_a$ during hibernation bouts with a time lag due to thermal inertia of the animals. The biggest difference between $T_b$ and $T_a$ occurred at the beginning of both the nocturnal and diurnal phase because of the rapidly changing $T_a$ at those times. The difference diminished during the course of nocturnal and diurnal phase and was lowest towards the end of each phase. Because of changing $T_a$, average hibernation $T_b$ (17.7 ± 0.6 °C; N=12, n=42) was slightly lower than average $T_a$, which was 18.5 ± 0.5 °C (*t-test*, $t = 5.112$, $p < 0.001$). No such

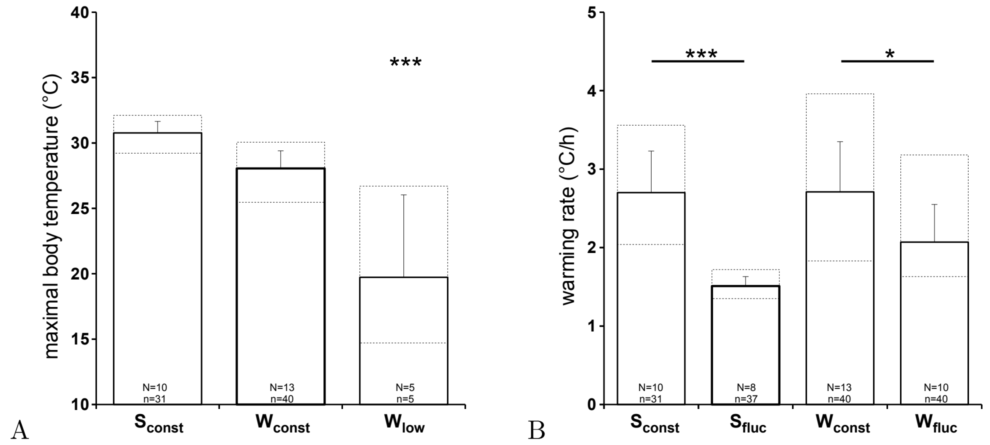

Figure 3.2.7: Maximal arousal body temperature and warming rates for all constant ambient temperature conditions

Maximal $T_b$ during arousals and activity phases (A) and warming rates from onset of arousal/activity phase till reaching maximum $T_b$ (B) for $W_{low}$, $W_{const}$ and $S_{const}$. Values are means, whiskers indicate the standard deviation and dashed boxes indicate the range (*** = p<0.001, ** = p<0.01, * = p<0.05)

difference was found between minimum hibernation $T_b$ and minimum $T_a$ (*t-test*, $t = -1.565$, $p = 0.144$).

The maximal and average arousal $T_b$ was always higher than corresponding $T_a$ (see 3.2.5B). Average and maximal $T_b$ for $W_{fluc}$ were 23.8 ± 1.3 °C (N=10, n=40) and 29.5 ± 1.1 °C (N=10, n=40), respectively. The highest observed $T_b$ of any animal for $W_{fluc}$ was 31.4 °C. Maximal arousal $T_b$ of $W_{fluc}$ did not differ compared to $S_{fluc}$ (*ANOVA*, $F = 22.977$, $p < 0.001$; *Bonferroni*, $p = 1$), nor did it differ from that observed in $W_{const}$ (*ANOVA*, $F = 22.977$, $p < 0.001$; *Bonferroni*, $p = 1$). The majority of arousal $T_b$ maxima occurred one to three hours before dusk (15:00 to 17:00 hours) although some also occurred during the first half of the night (fig. 3.1.4D). Warming rate of $W_{fluc}$ was 2.1 ± 0.5 °C/h (N=10, n=40) and was lower compared to $W_{const}$ (*ANOVA*, $F = 11.516$, $p < 0.001$; *Bonferroni*, $p = 0.033$) but did not differ compared to that observed in $S_{fluc}$ (*ANOVA*, $F = 11.516$, $p < 0.001$; *Bonferroni*, $p = 0.231$).

### 3.2.5 Oxygen consumption

Due to technical problems, no $VO_2$ data are available for the $W_{low}$ condition.

#### 3.2.5.1 Constant ambient temperature condition $W_{const}$

During hibernation bouts, $VO_2$ averaged 0.06 ± 0.02 $ml_{O_2}g^{-1}h^{-1}$ (N=4, n=12; fig. 3.2.8A), which was lower than $VO_2$ during daily torpor bouts during $S_{const}$ (*ANOVA*, $F = 11.396$,

Table 3.2: Oxygen consumption and percentage of energy saving compared to normothermy for ambient temperature conditions $W_{const}$ and $W_{fluc}$

| | $W_{const}$ | | | $W_{fluc}$ | | |
|---|---|---|---|---|---|---|
| | mean ± SD | energy saving | | mean ± SD | energy saving | |
| normothermy | 0.56 ± 0.24 | - | - | 0.56 ± 0.24 | - | - |
| daily average | 0.18 ± 0.09 | 68 % | * | 0.12 ± 0.04 | 79 % | *** |
| arousal | 0.79 ± 0.12 | -36 % | n.s. | 0.26 ± 0.11 | 54 % | *** |
| hibernation bout | 0.06 ± 0.02 | 89 % | ** | 0.10 ± 0.04 | 82 % | *** |

Oxygen consumption is given in $ml_{O_2}g^{-1}h^{-1}$, normothermy values are an average of $S_{const}$ and $S_{fluc}$

$p < 0.001$; *Bonferroni*, $p < 0.001$). Minimum $VO_2$ during hibernation bouts was 0.03 ± 0.01 $ml_{O_2}g^{-1}h^{-1}$ (N=4, n=12). When the animal aroused, $VO_2$ increased over 20-fold to 0.79 ± 0.12 $ml_{O_2}g^{-1}h^{-1}$ (N=4, n=8) as shown in figure 3.2.8B (*t-test*, $t$ = 14.681, $p < 0.001$) and maximal arousal $VO_2$ averaged 1.73 ± 0.20 $ml_{O_2}g^{-1}h^{-1}$ (N=4, n=8), with the highest observed value reaching 2.16 $ml_{O_2}g^{-1}h^{-1}$. Maximal arousal $VO_2$ did not differ compared to maximal activity phase $VO_2$ of $S_{const}$ (*ANOVA*, $F$ = 3.490, $p$ = 0.029; *Bonferroni*, $p$ = 0.565) and neither did average arousal $VO_2$ (*ANOVA*, $F$ = 16.096, $p < 0.001$; *Bonferroni*, $p$ = 0.060). Oxygen consumption during the warming phase was 1.11 ± 0.23 $ml_{O_2}g^{-1}h^{-1}$ (N=4, n=8; fig. 3.2.8C) and did not differ compared to $S_{const}$ (*ANOVA*, $F$ = 19.003, $p < 0.001$; *Bonferroni*, $p$ = 0.114). Average daily $VO_2$ was 0.18 ± 0.09 $ml_{O_2}g^{-1}h^{-1}$ (N=4; fig. 3.2.8D), which equated to a DEE of 0.088 ± 0.043 $kJd^{-1}g^{-1}$ (N=4) and was not different from daily $VO_2$ of $S_{const}$ (*ANOVA*, $F$ = 8.099, $p$ = 0.001; *Bonferroni*, $p$ = 0.062). The energy savings compared to normothermy (calculated from $S_{const}$ and $S_{fluc}$) are described in table 3.2 (*ANOVA*, $F$ = 13.281, $p < 0.001$; *Bonferroni* see table).

The pattern of $VO_2$ as a function of $T_b$ is shown in figure 3.2.9A. Oxygen consumption rose quickly and remained elevated over a broad range of $T_b$, from around 18 to 28 °C (red line). When $T_b$ declined again, $VO_2$ decreased rapidly to low levels (black line).

#### 3.2.5.2 Fluctuating ambient temperature condition $W_{fluc}$

Oxygen consumption during hibernation bouts during $W_{fluc}$ was 0.10 ± 0.04 $ml_{O_2}g^{-1}h^{-1}$ (N=12, n=42; fig. 3.2.8A) and did not differ compared to that observed during $W_{const}$ (*ANOVA*, $F$ = 11.396, $p < 0.001$; *Bonferroni*, $p$ = 0.935) nor during $S_{fluc}$ (*ANOVA*, $F$ = 11.396, $p < 0.001$; *Bonferroni*, $p$ = 1). Dividing the hibernation bout $VO_2$ between the nocturnal and diurnal phase showed that the values were lower during the nocturnal phase (0.06 ± 0.02 $ml_{O_2}g^{-1}h^{-1}$; N=12, n=42) compared to the diurnal phase (0.13 ± 0.06 $ml_{O_2}g^{-1}h^{-1}$; N=12, n=42; *t-test*, $t$ = 3.921, $p$ = 0.002). This relationship is shown in figure 3.2.10A, that shows $VO_2$ versus $T_b$ during hibernation. It can be seen that $VO_2$ stays constant at low $T_b$, but rises with elevating $T_b$. The minimum values during hibernation were 0.03 ± 0.01 $ml_{O_2}g^{-1}h^{-1}$ (N=12, n=42). A

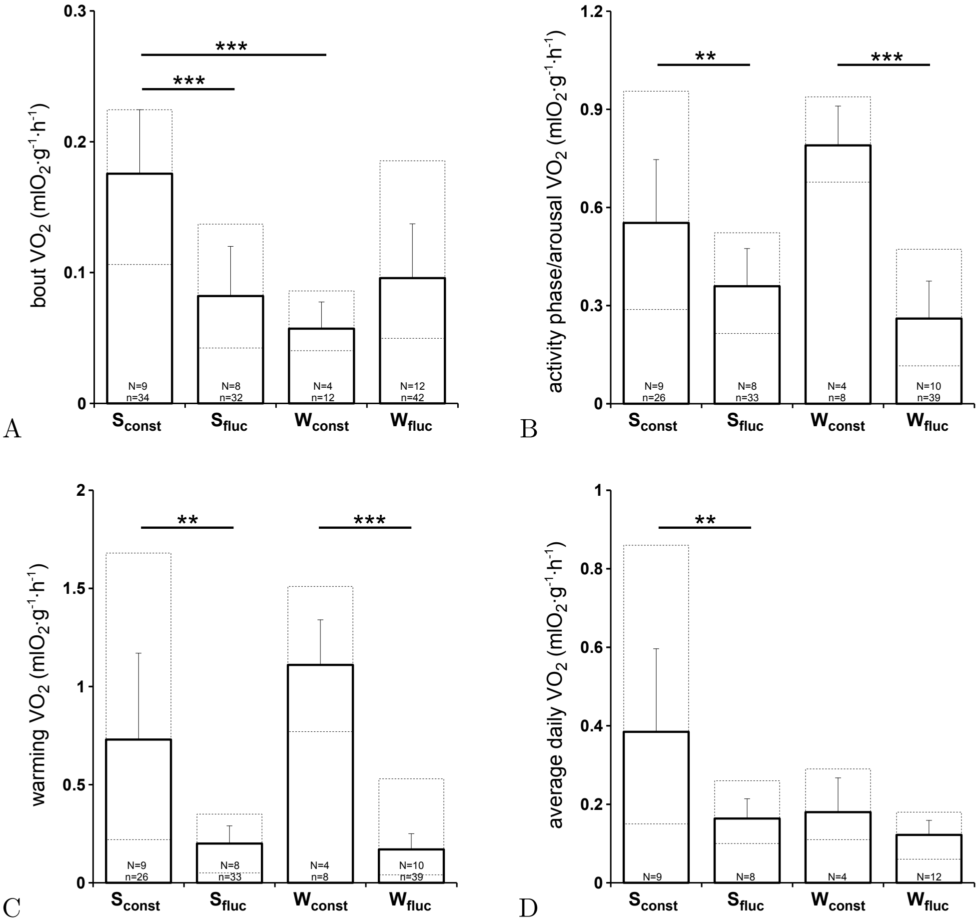

Figure 3.2.8: Oxygen consumption values for all conditions

Average $VO_2$ for daily torpor/hibernation bouts (A), average $VO_2$ for activity phases/arousals (B), warming $VO_2$ during activity phases/arousals (C) and average daily $VO_2$ (D) for the winter conditions $W_{fluc}$ and $W_{const}$, and for the summer conditions $S_{fluc}$ and $S_{const}$. Values are means, whiskers indicate the standard deviation and dashed boxes indicate the range ($*** = p < 0.001$, $** = p < 0.01$, $* = p < 0.05$)

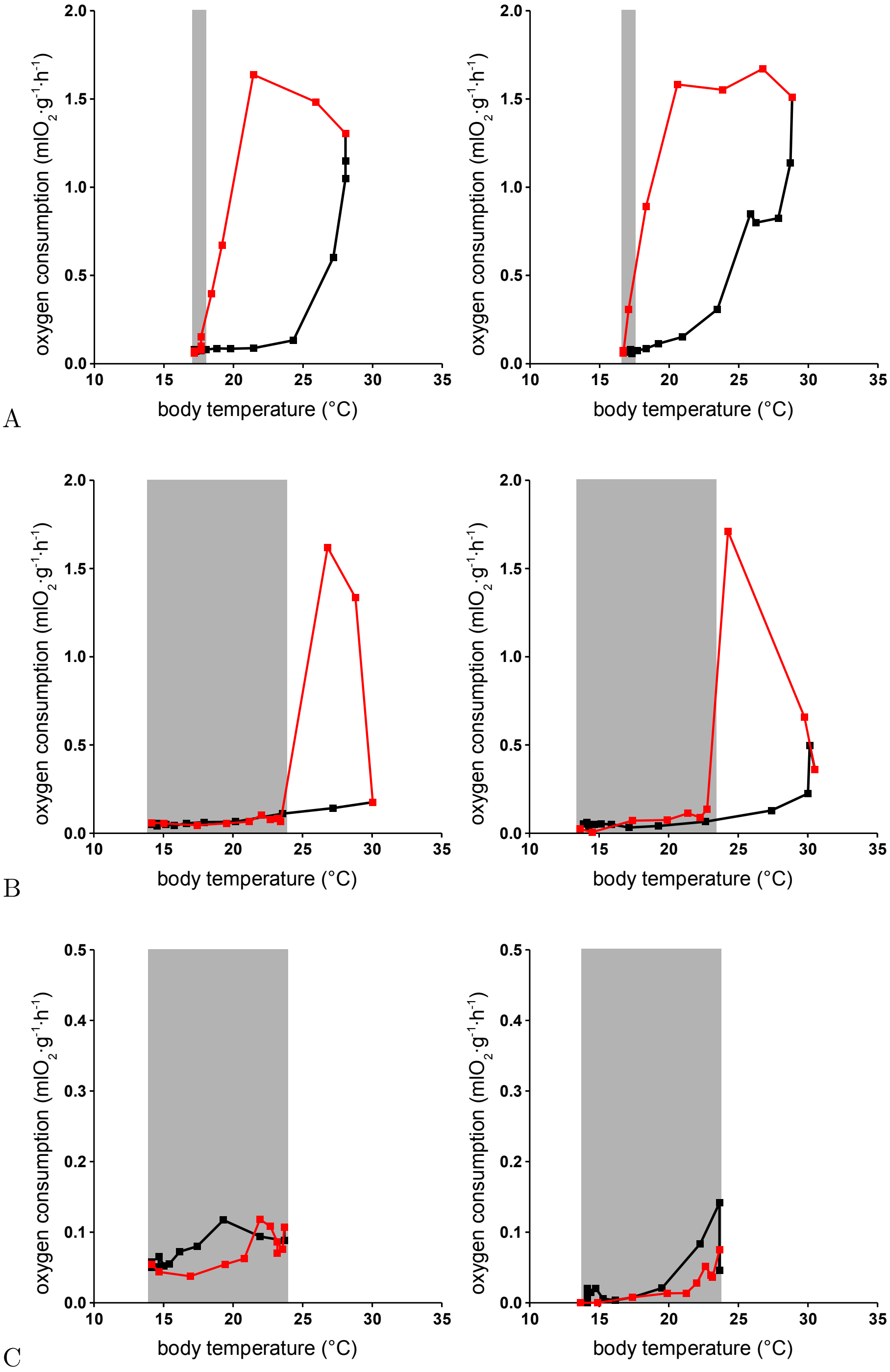

Figure 3.2.9: Oxygen consumption as a function of body temperature during the winter conditions $W_{const}$ and $W_{fluc}$

Oxygen consumption over a 24 hour period plotted against corresponding $T_b$ for $S_{const}$ (A; left: animal F0501, right: animal M0501) and $S_{fluc}$ (B; left: animal F0402, right: animal F0508), both including an arousal and parts of a hibernation bout, and for $S_{fluc}$ during a hibernation bout only (C; left: animal F0402, right: animal F0508). Red lines and squares are active and passive warming (start to $T_b$ maximum), black lines and squares are cooling ($T_b$ maximum to end). Shaded areas indicate the $T_a$ range over the displayed 24 hour period. Note the different scale of the y-axis in part C.

2.5-fold increase was noted for the arousal, yielding 0.26 ± 0.11 $ml_{O_2}g^{-1}h^{-1}$ (N=10, n=39*; t-test*, $t = 6.064$, $p < 0.001$), with an average maximal arousal $VO_2$ of 0.90 ± 0.55 $ml_{O_2}g^{-1}h^{-1}$ (N=10, n=39) and the highest observed arousal $VO_2$ was 3.09 $ml_{O_2}g^{-1}h^{-1}$ (see fig. 3.2.8B). Average and maximal arousal $VO_2$ were lower than for $W_{const}$ ($ANOVA$, $F = 16.096$, $p < 0.001$; *Bonferroni*, $p < 0.001$ for average arousal $VO_2$*;* $ANOVA$, $F = 3.490$, $p = 0.029$; *Bonferroni*, $p = 0.037$ for maximal arousal $VO_2$), but no difference was found compared to $S_{fluc}$ ($ANOVA$, $F = 16.096$, $p < 0.001$; *Bonferroni*, $p = 0.935$ for average arousal $VO_2$*;* $ANOVA$, $F = 3.490$, $p = 0.029$; *Bonferroni*, $p = 1$ for maximal arousal $VO_2$). Warming phase $VO_2$ averaged 0.17 ± 0.08 $ml_{O_2}g^{-1}h^{-1}$ (N=10, n=39; fig. 3.2.8C), which was lower compared to $W_{const}$ ($ANOVA$, $F = 19.003$, $p < 0.001$; *Bonferroni*, $p < 0.001$) but did not differ compared to $S_{fluc}$ ($ANOVA$, $F = 19.003$, $p < 0.001$; *Bonferroni*, $p = 1$). Daily $VO_2$ was 0.12 ± 0.04 $ml_{O_2}g^{-1}h^{-1}$ (N=10) which corresponded to a DEE of 0.060 ± 0.018 $kJd^{-1}g^{-1}$ (N=10). This did not differ from $W_{const}$ or $S_{fluc}$ ($ANOVA$, $F = 8.099$, $p = 0.001$; *Bonferroni*, $p = 1$ for $W_{const}$ and $S_{fluc}$). The energy savings compared to normothermy are described in table 3.2 ($ANOVA$, $F = 29.022$, $p < 0.001$; *Bonferroni* see table).

Figure 3.2.9B shows the pattern of $VO_2$ as a function of $T_b$ over a 24 h period including an arousal. Oxygen consumption stayed low over a wide range of $T_b$ (14 to 24 °C), indicating a reliance on passive heating (left part of red line). When the animals warmed above $T_a$, $VO_2$ rose quickly but declined rapidly as well after reaching the maximum $T_b$ (right part of red line). Part C of the figure shows two 24 h periods during hibernation with no arousal. Oxygen consumption remained low over the whole $T_b$ range (note the different scale of the y-axis compared to part A and B), but was nevertheless slightly correlated with $T_b$ (*linear regression*, $VO_2 = 0.011 * T_b - 0.113$, $R^2 = 0.307$, $F = 60.34$, $p < 0.001$).

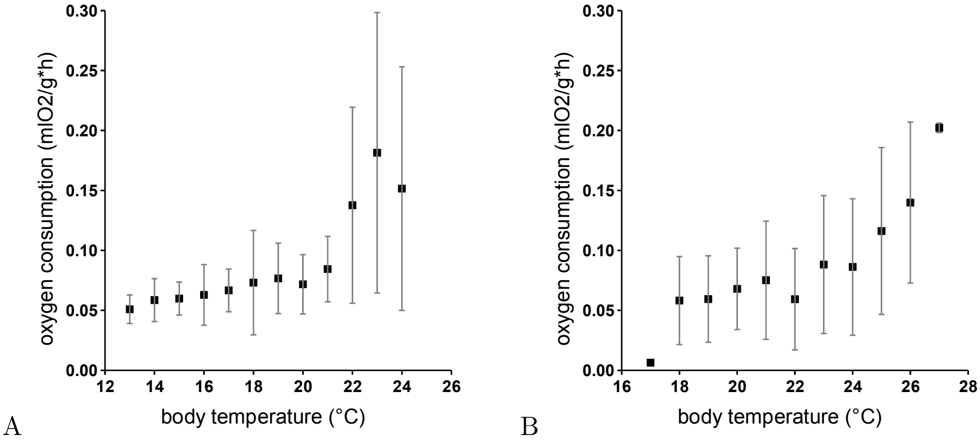

Figure 3.2.10: Oxygen consumption versus body temperature during hibernation and daily torpor bouts of $W_{fluc}$ and $S_{fluc}$

Hibernation $VO_2$ versus hibernation $T_b$ for $W_{fluc}$ (A) and $S_{fluc}$ (B). Error bars indicate the standard deviation.

Table 3.3: Summary of body temperature values and duration of hibernation/daily torpor bouts and arousals/activity phases

| | | hibernation / torpor bouts | | arousals / activity phases | | | |
|---|---|---|---|---|---|---|---|
| condition | | duration (days) | $T_b$ (°C) | duration (h) | $T_b$ (°C) | $T_b max$ (°C) | warming rate (°C/h) |
| $W_{low}$ | mean | 8.5 ± 3.1 | 12.3 ± 0.2 | 11:48 ± 3:57 | 17.4 ± 4.0 | 19.7 ± 6.3 | - |
| | range | 3.5 - 12.0 | 11.9 - 12.5 | 07:00 - 17:00 | 14.0 - 22.0 | 14.7 - 26.7 | - |
| | N, n | N=8, n=12 | N=8, n=12 | N=5, n=5 | N=5, n=5 | N=5, n=5 | - |
| $W_{const}$ | mean | 4.3 ± 2.2 | 17.6 ± 0.4 | 9:16 ± 2:21 | 24.5 ± 1.1 | 28.1 ± 1.4 | 2.71 ± 0.64 |
| | range | 0.6 - 8.0 | 16.53 - 18.32 | 03:00 - 15:00 | 21.6 - 26.1 | 23.5 - 30.1 | 1.48 - 3.96 |
| | N, n | N=16, n=45 | N=16, n=45 | N=13, n=40 | N=13, n=40 | N=13, n=40 | N=13, n=40 |
| $W_{fluc}$ | mean | 1.9 ± 1.4 | 17.7 ± 0.6 | 8:05 ± 1:09 | 23.8 ± 1.3 | 29.5 ± 1.1 | 2.07 ± 0.48 |
| | range | 0.3 - 4.6 | 15.8 - 19.2 | 04:00 - 12:00 | 19.5 - 25.8 | 24.7 - 31.4 | 1.31 - 3.18 |
| | N, n | N=12, n=42 | N=12, n=42 | N=10, n=40 | N=10, n=40 | N=10, n=40 | N=10, n=40 |
| $S_{const}$ | mean | 0.5 ± 0.2 | 24.6 ± 1.7 | 11:15 ± 0.35 | 28.5 ± 0.8 | 30.8 ± 0.9 | 2.70 ± 0.53 |
| | range | 0.1 - 0.9 | 22.0 - 27.4 | 03:00 - 44:00 | 24.9 - 30.2 | 29.0 - 32.8 | 1.78 - 5.37 |
| | N, n | N=10, n=41 | N=10, n=41 | N=10, n=31 | N=10, n=31 | N=10, n=31 | N=10, n=31 |
| $S_{fluc}$ | mean | 0.6 ± 0.0 | 21.1 ± 0.4 | 6:39 ± 0:36 | 28.4 ± 0.6 | 29.8 ± 1.0 | 1.51 ± 0.12 |
| | range | 0.5 - 1.0 | 19.4 - 23.6 | 04:00 - 08:00 | 27.0 - 29.9 | 27.9 - 31.7 | 0.97 - 2.03 |
| | N, n | N=8, n=36 | N=8, n=36 | N=8, n=37 | N=8, n=37 | N=8, n=37 | N=8, n=37 |

Values of the mean are given as mean ± SD

Table 3.4: Summary of oxygen consumption values

| condition | | daily $VO_2$ ($ml_{O_2}g^{-1}h^{-1}$) | hibernation / torpor bouts: $VO_2$ ($ml_{O_2}g^{-1}h^{-1}$) | hibernation / torpor bouts: minimal $VO_2$ ($ml_{O_2}g^{-1}h^{-1}$) | arousals / activity phases: $VO_2$ ($ml_{O_2}g^{-1}h^{-1}$) | arousals / activity phases: maximal $VO_2$ ($ml_{O_2}g^{-1}h^{-1}$) | arousals / activity phases: warming $VO_2$ ($ml_{O_2}g^{-1}h^{-1}$) |
|---|---|---|---|---|---|---|---|
| $W_{const}$ | mean | 0.18 ± 0.09 | 0.06 ± 0.02 | 0.03 ± 0.01 | 0.79 ± 0.12 | 1.73 ± 0.20 | 1.11 ± 0.23 |
| | range | 0.11 - 0.29 | 0.03 - 0.11 | 0.01 - 0.06 | 0.66 - 1.07 | 1.47 - 2.16 | 0.77 - 1.51 |
| | N, n | N=4 | N=4, n=12 | N=4, n=12 | N=4, n=8 | N=4, n=8 | N=4, n=8 |
| $W_{fluc}$ | mean | 0.12 ± 0.04 | 0.10 ± 0.04 | 0.02 ± 0.02 | 0.26 ± 0.11 | 0.90 ± 0.55 | 0.17 ± 0.08 |
| | range | 0.06 - 0.18 | 0.04 - 0.19 | 0.01 - 0.05 | 0.08 - 0.63 | 0.11 - 3.09 | 0.04 - 0.53 |
| | N, n | N=10 | N=12, n=42 | N=12, n=42 | N=10, n=39 | N=10, n=39 | N=10, n=39 |
| $S_{const}$ | mean | 0.38 ± 0.21 | 0.18 ± 0.05 | 0.12 ± 0.05 | 0.55 ± 0.19 | 1.24 ± 0.55 | 0.73 ± 0.44 |
| | range | 0.15 - 0.86 | 0.07 - 0.38 | 0.01 - 0.26 | 0.17 - 1.05 | 0.32 - 3.43 | 0.22 - 1.68 |
| | N, n | N=9 | N=9, n=34 | N=9, n=34 | N=9, n=26 | N=9, n=26 | N=9, n=26 |
| $S_{fluc}$ | mean | 0.16 ± 0.05 | 0.08 ± 0.04 | 0.03 ± 0.03 | 0.36 ± 0.12 | 0.95 ± 0.28 | 0.20 ± 0.09 |
| | range | 0.10 - 0-26 | 0.04 - 0.25 | 0.01 - 0.14 | 0.10 - 0.59 | 0.21 - 1.69 | 0.05 - 0.35 |
| | N, n | N=8 | N=8, n=32 | N=8, n=32 | N=8, n=33 | N=8, n=33 | N=8, n=33 |

Values of the mean are given as mean ± SD

Table 3.5: Summary of statistics

| | hibernation / torpor bouts | | | | arousals / activity phases | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| comparison | duration | $T_b$ | $VO_2$ | $VO_2min$ | duration | $T_b$ | $T_bmax$ | warming rate | $VO_2$ | $VO_2max$ | warming $VO_2$ | daily $VO_2$ |
| $W_{low}$-$W_{const}$ | *** | *** | - | - | n.s. | *** | *** | - | - | - | - | - |
| $W_{low}$-$S_{const}$ | *** | *** | - | - | n.s. | *** | *** | - | - | - | - | - |
| $W_{const}$-$S_{const}$ | *** | *** | *** | *** | n.s. | *** | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. |
| $W_{fluc}$-$S_{fluc}$ | n.s. | *** | n.s. | n.s. | n.s. | *** | n.s. | n.s. | n.s. | n.s. | n.s. | n.s. |
| $W_{const}$-$W_{fluc}$ | * | *** | n.s. | n.s. | n.s. | n.s. | n.s. | * | *** | * | *** | n.s. |
| $S_{const}$-$S_{fluc}$ | n.s. | *** | *** | *** | n.s. | n.s. | n.s. | *** | ** | n.s. | ** | ** |

$*** = p < 0.001$, $** = p < 0.01$, $* = p < 0.05$, n.s. = not significant

# Chapter 4

# Discussion

## 4.1 Methodological considerations

### 4.1.1 Definition of daily torpor and hibernation bouts

Although it seems obvious that hibernation bouts and arousals or daily torpor bouts and activity phases are different physiological states, the distinction between these remains arbitrary. Different definitions of what constitutes a daily torpor or hibernation bout are found in the literature. An early definition was that of Hudson (1978), who suggested that the torpid state was reached when $T_b$ falls below 31 °C. Criteria that are based on the animal reaching a particular set temperature have been used in numerous studies (see Barclay et al. (2001) for an exhaustive review). However, such an approach disregards the fact that different species have different normothermic $T_b$. A fall from a normothermic $T_b$ of 40 °C, e.g. birds, to $<$ 31°C cannot be compared to a fall from a normothermic $T_b$ of 33 °C as found in some marsupials.

Therefore, Barclay et al. (2001) proposed that once normothermic $T_b$ is known, torpor should be defined as $T_b$ below that level. This should be done at the least for the species, but far better for individuals. Although this approach controls for variation at the species and individual level, it is still not a useful definition for the work with *E. telfairi*. The lesser hedgehog tenrec relies entirely on daily torpor and hibernation and does not show prolonged phases of normothermy. Thus it is very difficult to obtain a credible estimate of normothermic $T_b$ in this species.

The definition of normothermy used in the present study ($T_b > 28°C$) is an arbitrary one and was chosen to gain an estimate of normothermic $VO_2$. Nonetheless, this definition of normothermy is unsuitable for discrimination between daily torpor/hibernation bouts and activity phases/arousals, particularly since maximum $T_b$ during activity phases and arousals decreases with decreasing $T_a$ and sometimes did not exceed 28°C during the $W_{low}$ experimental condition. To obtain a working definition for all experimental conditions, a different approach was taken. The animals were regarded as torpid when $T_b$ was at or only slightly above $T_a$ ($T_b - T_a \leq 2°C$). Similar definitions had been used successfully by other researchers (e.g. Arlettaz et al., 2000;

Hickey and Fenton, 1996). Although this is workable, such a definition has some drawbacks. The difference between $T_b$ and $T_a$ decreases at high $T_a$ and when $T_a$ approaches normothermic $T_b$ this method no longer works. Nevertheless, the definition remained useful within the range of $T_a$ employed in this study.

### 4.1.2 Body temperature measurements

The most accurate and reliable method to measure $T_b$ in normothermic, as well as in hibernating animals is to implant temperature-sensitive data loggers into the abdominal cavity of the animals. But this requires anaesthesia and surgery, both potentially harmful for the animals. It was thus decided to measure $T_b$ with externally attached temperature loggers (iButtons), that measure skin temperature rather than $T_b$. That externally attached devices can deliver accurate data has been shown on many occasions. A comparison of rectal versus skin temperature in big brown bats *Eptesicus fuscus* at $T_a$ between 2 °C and 26 °C showed that skin temperature was a good estimate of rectal temperature in torpid and active animals, although there was a slight effect of $T_a$ on skin temperature (Barclay et al., 1996). In that study skin temperature was never $> 3.3$ °C below rectal temperature and even small decreases in $T_b$ could be accurately inferred from skin temperatures. Similar findings have been reported for the bat *Carollia perspicillata* (Audet and Thomas, 1996). Dausmann (2005) compared data from external collar temperature transmitters to that from implanted temperature-loggers in the fat-tailed dwarf lemur *Cheirogaleus medius.* She found that temperature data from collar transmitters can be used to estimate core $T_b$ over a wide range of $T_a$ and concluded that external devices are adequate for determining general patterns of $T_b$. This applies particularly to small animals that sleep or hibernate in a curled up position, like *E. telfairi.*

Scholl (1974) measured $T_b$ in lesser hedgehog tenrecs with a monitor attached to the skin. He reported skin temperatures during activity phases of around 30 °C and the highest observed $T_b$ was slightly below 33°C. This corresponds with the values observed during my study. In a recent study temperature loggers were implanted into three lesser hedgehog tenrecs (Lovegrove and Genin, 2008). That study was under semi-natural conditions wherein $T_a$ fluctuated between 19.6 °C and 31.1 °C diurnally. They found a mean maximum $T_b$ of 30.7 °C and the highest individual $T_b$ was 36.2 °C. While their mean maximum $T_b$ is similar to the mean maximum $T_b$ in my study, the highest individual $T_b$ observed by Lovegrove and Genin is considerably higher. If this is caused by the different $T_b$ measurement method or by the higher $T_a$ remains unclear. Nevertheless, the similarity of the observed values is a good indication of the reliability and accuracy of externally attached temperature loggers. The accuracy will be even higher when the animals are in torpor or hibernation because they sleep curled up into a ball thereby ensuring even better contact between skin and data logger.

Taping temperature loggers to the animal's belly proved to be the best solution but it remained difficult to maintain consistent adhesion between the monitor and the skin. When temperature loggers are implanted into the abdomen or under the skin, they remain permanently in place. However, if the loggers are just taped to the shaved skin, they will drop off eventually and the degree of adhesion needs to be monitored regularly.

Handling of the animals, i.e. picking them up and checking the taped loggers, did not seem to effect the animals' behaviour or $T_b$. They continued with their behaviour after being put down, regardless of being active or in hibernation. Adhesion of the logger and tape during the hibernation period, when the animals were inactive for most of the time, was excellent and only rarely did the loggers require attention. During summer, when the animals were active and running through the cage, the loggers became unstuck after three to four days and had to be reinstalled. Nonetheless, doing so did not appear to interfere with their normal behaviour.

### 4.1.3 Measurement of energy consumption

Energy expenditure can be measured by direct calorimetry since all energy consumed is ultimately released as heat. But this method has many drawbacks and is not suitable for long-term measurements, as were required in this study. Hence another, indirect method had to be used to measure energy consumption.

Indirect calorimetry measures turnover rates of substances and products involved in metabolism. There are two methods that are routinely used, the doubly-labelled water method and measurement of respiratory gases. While the doubly-labelled water method is suitable for long-term measurements, it yields only an overall energy budget over the whole measurement and does not allow for any temporal resolution (Butler et al., 2004).

The analysis of respiratory gases, namely oxygen and carbon dioxide, has developed as the gold standard for measurements of metabolic rate when temporal resolution is required (Walsberg and Hoffman, 2005). It has been widely used in hibernation studies as well. In the present study an open-circuit respirometry system was used because it allowed us to use the animals' nestboxes as respirometry chambers. This way they could remain in a familiar environment at least in part. That animals could leave the nestboxes on their on volition during the experiments rendered their natural behaviour possible but it also decreased the reliability of $VO_2$ data. I did not track when the animals left and entered the nestboxes. During the winter the animals were inactive and spent nearly the entire time in the nestboxes. The animals stayed in the nestboxes during most arousals and if they left the nestboxes, they did so only for a short time, i.e. a few minutes, to drink or urinate. Therefore, $VO_2$ measurements during winter should be accurate.

Summer measurements, on the other hand, most likely underestimate real $VO_2$. The animals were active throughout summer and entered and left their nestboxes frequently. Although data was excluded from analysis, when $VO_2$ fell to zero, indicating that the animals were not inside the nestbox, the time shortly after an animal left or entered the nestbox did not reflect real $VO_2$ since a new steady state within the nestbox had to be established. The available data cannot identify whether an animal left the nestbox only for a brief moment or left and entered it multiple times in close succession. Furthermore, the highest rate of $VO_2$ most likely occurred outside the nestbox, when the animals were running and climbing inside the cage. Maximal $VO_2$

can increase by 10-fold the BMR and even during moderate activity, $VO_2$ increases by three to four fold the BMR (Heldmaier, 2003). Oxygen consumption during the activity phase during summer is therefore most likely underestimated. Energy consumption during daily torpor bouts, when the animals were resting inside the nestbox, however, should be accurate.

## 4.2 Daily torpor during summer

Daily torpor is found in a wide range of mammalian orders, including marsupials, rodents, elephant shrews, primates, colugos, bats, insectivores and even-toed ungulates (Heldmaier et al., 2004). These animals represent a wide range of taxonomic as well as habitat and climatic affinities. Subsequently, torpor pattern, use and function differ between species and habitats.

### 4.2.1 Daily torpor pattern

Most species are facultative users of daily torpor, using it at certain times within a seasonal cycle or sporadically if needed. If daily torpor is predominantly used during one season of the year, it is normally the cold season, when $T_a$ is low and food resources are scarce (Geiser, 2004). This is understandable since this hypometabolic state is primarily used for energy conservation (Boyer and Barnes, 1999). In other habitats, times with low food availability might coincide with the warm season, making this the daily torpor season. This is the case for the subtropical blossom-bat *Syconycteris australis* that feeds on nectar and becomes torpid in the summer when nectar availability in its habitat is low (Coburn and Geiser, 1998). In contrast to temperate and arctic regions, where energy conservation is the predominant function of daily torpor, subtropical and tropical species benefit from saving water during daily torpor (Cryan and Wolf, 2003; Cooper et al., 2005; Schmid and Speakman, 2000). The gerbil *Gerbillus pusillus* enters torpor not only in response to food deprivation, but also in response to water deprivation (Buffenstein, 1985) and the mouse lemur *Microcebus murinus* has lower field water turnover rates when using torpor compared to normothermic individuals (Schmid and Speakman, 2000).

*E. telfairi* appears to be obliged to use torpor every day during summer. Animals entered torpor every single day measured, except for one female that remained normothermic for two days on two occasions. This is similar to the findings of other investigators, who also observed torpor in this species on every day that they were studied (Lovegrove and Genin, 2008; Scholl, 1974). There are only few reports of other species that rely similarly on daily torpor. Although the lesser long-eared bat *Nyctophilus geoffroyi* enters torpor every day in the field during summer (Turbill et al., 2003a,b), there are indications that this species is capable of longer stretches of normothermy (Geiser and Brigham, 2000).

Although both sexes of lesser hedgehog tenrecs do not lack the ability to maintain a high $T_b$ for longer periods on principle, only females are known to elevate their $T_b$ during both pregnancy and lactation to about 32°C to 34°C (Poppitt et al., 1994). The female in the present study was neither pregnant nor lactating, presenting the first evidence of extended normothermy outside gestation and lactation in this species. While female lesser hedgehog tenrecs are evidently capable of longer stretches of normothermy, this has never been observed in males. Heterothermy in general is widespread amongst the Tenrecidae. Torpor or hibernation are found in spiny tenrecs (Lovegrove and Genin, 2008; Nicoll, 1986; Buffenstein and Salton, 2003), large-eared tenrecs (Stephenson and Racey, 1993a) and furred tenrecs (Stephenson, 1994; Stephenson and Racey, 1993b). Otter shrews have yet to be investigated. Besides being heterothermic, many species within the Tenrecidae also show limited or poorly developed thermoregulation, but can elevate their $T_b$ and keep it relatively constant, i.e. homeothermic, during pregnancy and lactation (Racey and Stephenson, 1996).

Evolutionary, *E. telfairi* could have either lost or never developed the ability to maintain extended periods of elevated $T_b$. If the species lost this ability and as a result obligate heterothermy is a derived trait, what environmental conditions would favour the loss of homeothermy? No factor comes to mind immediately. The ability to employ torpor to minimize energy consumption under unfavourable conditions, i.e. bad weather or food and water shortages, is surely a major selective advantage. But being unable to maintain elevated $T_b$ and extend foraging or mating under highly favourable conditions, when food and other resources are plentiful, does not seem to have any plausible benefit. Torpor is a viable adaptation to lower the risk of extinction in highly variable and quickly changing environments (Geiser and Turbill, 2009; Liow et al., 2009), but this occurs in both good and bad environmental conditions. It seems unlikely that Madagascar's climate promoted the loss of homeothermy since no other Madagascan mammal thus far investigated behaves similarly. The division of the Madagascan climate into a dry and wet season (Jury, 2004) surely fostered the development of hibernation during the unfavourable dry season in some indigenous mammals, including *E. telfairi*, but this cannot account for *E. telfairi*'s thermogenic limitations and neither can factors associated with their habitat or diet.

It is therefore likely that the obligatory heterothermy and poor thermogenetic capacity found in *E. telfairi* is an ancestral trait. The occurrence of torpor in many different mammalian clades speaks strongly for a plesiomorphic origin of heterothermy (Lovegrove et al., 1999a) and as Grigg et al. (2004) argue, homeothermic endothermy evolved likely via heterothermy. From this perspective, *E. telfairi* can be considered a proto-endotherm, displaying the most primitive form of heterothermy (Lovegrove and Genin, 2008). How endothermy has evolved is still under intense debate and different concepts have been brought forward (Koteja, 2004). Apart from the popular aerobic capacity model (Bennett and Ruben, 1979) that explains endothermy as a by-product of maintaining a high level of perpetual locomotor activity, some newer models highlight the potential benefit of (intermittent) endothermy for parental care (Farmer, 2000; Koteja, 2000). Higher $T_b$ can accelerate incubation and growth of the young and also enhance energy assimilation needed for lactation.

*E. telfairi* meets many of the criteria for a hypothetical, evolutionarily early species that can elevate its $T_b$ briefly only during the activity phase and during the gestation period. The short-beaked echidna *Tachyglossus aculeatus* is another animal that is considered a proto-endotherm, displaying primitive traits such as a strikingly heterothermic $T_b$ pattern most of the time and shivering thermogenesis (Grigg et al., 2004). Similar to *E. telfairi* during gestation and lactation, short-beaked echidnas possess the capacity for homeothermic endothermy during incubation of their eggs (Beard and Grigg, 2000). Thus it can be assumed that the lack of normothermy in the lesser hedgehog tenrec is an ancestral trait, which, with its position within the basal afrotherian clade, argues for plesiomorphy of heterothermy in mammals.

### 4.2.2 Energy savings

Daily torpor is a very effective mechanism for saving energy. Metabolic rate during daily torpor bouts can be reduced by over 90%, yielding daily energy savings of 20-50% (Geiser and Ruf, 1995). The tropical hibernator *E. telfairi* lowered its MR by approximately 80% compared to normothermy, which is a considerable reduction given the high $T_a$ and consequently high $T_b$ during daily torpor bouts. Calculation of daily energy savings is not possible in the lesser hedgehog tenrec because it enters daily torpor every day. However, it is possible to compare the $VO_2$ of the female that remained normothermic for more than 24 h on two occasions, although $VO_2$ data is just available for one episode. During the normothermic 24 h period this animal consumed 0.96 $ml_{O_2}g^{-1}h^{-1}$ compared to 0.65 $ml_{O_2}g^{-1}h^{-1}$ on a daily torpor day. This suggests a daily energy saving of around 30%, whereas the significance of this calculation is doubtful due to the lack of longer normothermic periods in *E. telfairi*.

Nectar feeding bats of the species *Glossophaga soricina*, that enter torpor in response to energetic restrictions, lower their torpid MR to 5% of normothermic MR (Kelm and von Helversen, 2007). Another tropical bat, the northern blossom-bat *Macroglossus minimus*, can reduce its MR by 60-80% (Bartels et al., 1998). Although Madagascar's lemurs do not use daily torpor during the wet season as *E. telfairi* does, some become torpid during the dry season in the winter. The pygmy mouse lemur *Microcebus myoxinus* uses daily torpor during the dry season and reduces its MR during torpor by 86% compared to normothermic RMR, yielding DEE reductions of 40% (Schmid et al., 2000). Similar reductions have been found in the gray mouse lemur *M. murinus* (Schmid, 1998). Compared to these tropical mammals, the observed MR reduction of 80% in *E. telfairi* is of the same magnitude, suggesting that daily torpor in the tropics is of the same energetic significance in different animals. Furthermore, it shows that tropical daily torpor can be as efficient as daily torpor in temperate and arctic regions.

### 4.2.3 Effects of fluctuating ambient temperature

There are few natural habitats that experience little or no fluctuations in diurnal $T_a$. Among these are marine and limnic habitats, that are well buffered by the high specific heat capacity of water, as well as caves and deep burrows. All other habitats are exposed to fluctuations in $T_a$ sometimes of considerable magnitude. Therefore, an experimental regime of fluctuating $T_a$ reflects the natural habitat of most mammals more accurately than a regime of constant $T_a$.

The lesser hedgehog tenrec, as an inhabitant of south-western Madagascar is exposed to diurnal $T_a$ fluctuations of more than 10°C (Jury, 2004; Lovegrove and Genin, 2008). The experimental summer condition $S_{fluc}$ imitates its natural habitat as it mimics the diurnal $T_a$ fluctuations *E. telfairi* is exposed to. An experimental regime with fluctuating $T_a$ allows an investigation of how the lesser hedgehog tenrec copes with similar fluctuations in its natural habitat. Furthermore, such a regime enables insights into if and how these animals utilize daily $T_a$ fluctuations for passive warming and if fluctuations in $T_a$ affect the pattern of hibernation. A constant $T_a$ regime is, in effect, an artificial condition that would never be encountered by a hedgehog tenrec in the wild, unless it spends extended periods of time in a well-buffered place, such as an underground burrow or a deep tree hollow. Nonetheless, including this as an experimental treatment serves to demonstrate *E. telfairi*'s thermogenic capacities and daily torpor pattern in the absence of passive warming.

Maximal $T_b$ during activity phases did not differ between the two summer conditions and was of the same magnitude as that reported for intraperitoneally-implanted temperature loggers by Lovegrove and Genin (2008). This is a good indication of the validity of our methods as discussed in section 4.1.2.

Fluctuating $T_a$ influenced the timing of activity phases. As can be seen in the circular plots (see fig. 3.1.4), maximal $T_b$, which acts as an indicator of the most active time of the activity phase, is better synchronised during the fluctuating $T_a$ regime than the constant $T_a$ regime. As *E. telfairi* follows a diel activity cycle, this must be triggered and synchronised by external *zeitgebers*, especially since timing of daily torpor is importance for its optimal effectiveness (Körtner and Geiser, 2000). Although photoperiod has a pronounced effect on the timing of daily torpor (Heldmaier et al., 1982, 1989), there are some species (e.g. the small marsupial dunnarts) that are insensitive to photoperiodic cues (Holloway and Geiser, 1996). In the stripe-faced dunnart, *Sminthopsis macroura,* daily fluctuations in $T_a$ act as weak *zeitgebers* (Francis and Coleman, 1990). The results of the present study suggest that fluctuating $T_a$ is an effective *zeitgeber* for the timing of the activity phase in the lesser hedgehog tenrec.

Although the duration of the activity phase for $S_{fluc}$ of around 7 h was lower compared to 11 h for $S_{const}$, this was not statistically significant. The difference could be caused by the different definition of activity phases and daily torpor bouts between fluctuating and constant $T_a$ conditions. But the time spent normothermic within the activity phases was reduced by nearly 50% during $S_{fluc}$ compared to $S_{const}$. The reason for this reduction in normothermic time is unclear. Energetically, *E. telfairi* could afford to stay normothermic in fluctuating $T_a$ conditions for longer because less energy is used for warming. That the observed response is reversed could indicate that the *zeitgeber* function of fluctuating $T_a$ enables a more "orderly" daily torpor pattern. During constant $T_a$ the thermal stimuli that synchronize the circadian clock are absent. This could not only affect the timing of the activity phase and maximal $T_b$, but also the duration of the normothermic period.

While there was no effect of fluctuating $T_a$ on $T_b$ and duration of torpor bouts, $VO_2$ was strongly affected. Oxygen consumption during daily torpor bouts for $S_{fluc}$ was less than half that for $S_{const}$, which is remarkable since there is no obvious reason for this reduction. A lower $T_a$ and the consequentially lower $T_b$ reduce $VO_2$ during hibernation bouts in the golden-mantled ground squirrel *Spermophilus lateralis*, demonstrating the temperature-dependence of $VO_2$ in the torpid state (Zimmer and Milsom, 2001). Lower $VO_2$ at lower $T_a$ during daily torpor bouts has also been found in the tropical primate *M. myoxinus* (Schmid et al., 2000). However, although $T_b$ during an average daily torpor bout under $S_{fluc}$ (21.1°C) was lower compared to $S_{const}$ (24.6°C), a $T_b$ reduction of 4°C is not sufficient to elicit such a big difference in $VO_2$. If the reduction of $VO_2$ was caused by temperature effects alone, this would imply an Q10 of over 10, which is considerably higher than the Q10s of 2 to 4 that are normally found in biological systems (Heldmaier, 2003). This indicates increased active metabolic inhibition at lower $T_b$. However, it is unclear why *E. telfairi* should show a great downregulation of its metabolism at lower $T_b$ compared to higher $T_b$ during daily torpor bouts. The answer could lie in the more synchronized and orderly daily torpor pattern during fluctuating $T_a$, that allows *E. telfairi* to achieve deeper daily torpor bouts compared to constant $T_a$ conditions.

The lower energetic costs of the activity phase during fluctuating $T_a$ can be better explained. Warming up from the torpid state to normothermic $T_b$ is a costly process. During the hibernation season, most of the energy used is consumed for this process (Boyer and Barnes, 1999). Re-warming during daily torpor occurs each day and thus adds greatly to overall energy consumption compared to hibernation (Geiser and Ruf, 1995; Lovegrove et al., 1999b). Although MR during torpor can be as low as during hibernation (Lovegrove et al., 1999a; Wilz and Heldmaier, 2000), the frequent warming up increases the overall energy consumption considerable. Even small energy savings during re-warming will lead to considerable overall energy savings. Not surprisingly, many species use passive warming to reduce their warming costs by exploiting fluctuating $T_a$ or radiant heat (Schmid et al., 2000; Lovegrove et al., 1999b; Kobbe and Dausmann, 2009; Dausmann et al., 2005; Mzilikazi et al., 2002).

The lesser hedgehog tenrec can also use daily $T_a$ fluctuations very effectively. Warming costs, i.e. from the start of passive/active warming until reaching the normothermic $T_b$ ($> 28°C$), were reduced by nearly 75% during $S_{fluc}$ compared to $S_{const}$ (0.20 $ml_{O_2}g^{-1}h^{-1}$ vs. 0.73 $ml_{O_2}g^{-1}h^{-1}$). Overall this resulted in a reduction of daily energy requirements of about 60%. The difference between $T_b$ at the start of warming and the normothermic state $T_b$ was nearly twice that of $S_{fluc}$, but warming was effectively a passive process using the high $T_a$ (28.1°C). Although active warming was more costly, it was also faster. During active warming the animals reached normothermy after only 1.7 h at a warming rate of 2.7 °C/h compared to nearly 6 h at a rate of 1.5 °C/h during $S_{fluc}$. Nonetheless it remains uncertain whether the benefits of faster re-warming, e.g. extending foraging time, can offset the higher costs of warming in a natural setting.

The observed reduction in energy expenditure of *E. telfairi* during passive heating corresponds well with the findings of other authors. Schmid et al. (2000) calculated the potential energy savings during passive warming in the tropical pygmy mouse lemur *Microcebus myoxinus* to be around 60% compared to active warming. Reductions of comparable magnitude have been found in the stripe-faced dunnart *S. macroura*. Lovegrove et al. (1999b) reported energy savings of 65% when warming passively by a diel $T_a$ cycle (15°C-25°C). Energy savings in this Australian marsupial can even be as high as 85% when re-warming from torpor is assisted by radiant heat (Geiser and Drury, 2003). Basking has also been observed in another species of the genus *Sminthopsis*, the fat-tailed dunnart *S. crassicaudata*, where it is used for the reduction of energetic costs during warming as well (Warnecke et al., 2008). The benefit from passive heating, either by daily fluctuations of $T_a$ or by radiant heat, might lead to laboratory studies underestimating the energetic advantage of torpor in free-ranging mammals (Mzilikazi et al., 2002). Furthermore, the benefits of passive heating might explain why daily torpor is common in sunny regions and it indicates that daily torpor might occur more frequently at low latitudes than hitherto believed (Geiser and Drury, 2003).

## 4.3 Hibernation during winter

Two decades ago, hibernation was believed to occur exclusively in temperate and arctic mammals in order to cope with cold temperatures and food shortages during the winter. Only recently have mammals in the tropics be found to hibernate as well. With the advances in miniaturized temperature loggers, future field (and laboratory) studies are likely to reveal more tropical hibernators.

The Tenrecidae, as a family of tropical small to medium-sized mammals, were among the first tropical species in which hibernation was observed. The two species of streaked tenrecs *Hemicentetes nigriceps* and *H. semispinosus* hibernate during the austral winter (Stephenson and Racey, 1994) and the common tenrec *Tenrec ecaudatus* is also known to hibernate (Nicoll, 1985). Because of the widespread occurrence of hibernation in the subfamily Tenrecinae it is not surprising that *E. telfairi* is capable of hibernation, too.

### 4.3.1 Hibernation pattern

The lesser hedgehog tenrec shows a hibernation pattern comparable to that seen in arctic and temperate hibernators. Hibernation bouts of varying length are disrupted by periodic arousals. But despite this basic similarity, there are some key differences between hibernation patterns in tropical species and those observed in hibernators from colder climates.

Hibernation bouts are much longer in arctic and temperate species compared to the lesser hedgehog tenrec. The average duration of a hibernation bout in *E. telfairi* at 18°C was 4.3 d, with a longest duration of 8 d. This is considerably lower compared to arctic and temperate hibernators, in which an average hibernation bout might last for one to two weeks. Longer bouts of over a month are found in numerous species. The edible dormouse *Glis glis* can hibernate up to 34 d at a stretch without an arousal (Heldmaier, 2003) and even longer bouts of up to 45 d have been found in the European badger *Meles meles* (Fowler and Racey, 1988). The length and pattern of hibernation bouts of the only other tropical hibernators, the cheirogaleids, vary widely between different species. The gray mouse lemur *M. murinus* shows only short bouts with a maximum duration of 3.7 d (Schmid and Ganzhorn, 2009), whereas its close relative *M. griseorufus* has an average hibernation bout duration of 12 d and the longest hibernation bouts exceeded one month (Kobbe and Dausmann, 2009). However, only 20% of *M. griseorufus* hibernate, the other 80% use mainly daily torpor during the dry season. The longest continuous hibernation bouts of up to 70 d have been found in the fat-tailed dwarf lemur *C. medius*, but only if Ta exceeds 30°C regularly; if these conditions are not met, it uses shorter hibernation bouts with periodic arousals (Dausmann et al., 2005).

The duration of hibernation bouts varies not only between species, but is also influenced by dietary and ambient factors. Polyunsaturated fatty acids (PUFAs), or more specifically the ratio of n-6 to n-3 PUFAs, influence hibernation bout duration, likeliness of entering torpor and $T_b$ during hibernation. Higher ratios of n-6 to n-3 PUFAs increasing the length of hibernation bouts and the propensity to enter hibernation (Ruf and Arnold, 2008). Ambient temperature also affects the duration of hibernation bouts. Decreasing $T_a$ increased the length of hibernation bouts (French, 1982). A reduction in $T_a$ from 8°C to 2°C doubled the length of hibernation bouts in the golden-mantled ground squirrel *Spermophilus saturatus* (Geiser and Kenagy, 1988). A similar effect was observed in the present study, where duration of hibernation bouts of the lesser hedgehog tenrec increased from 4.3 d to 8.5 d with a reduction in $T_a$ from 18°C to 12°C. This suggests that the duration of hibernation bouts is temperature-dependent in tropical as well as temperate and arctic hibernators.

Although lower $T_a$ lengthens the duration of hibernation bouts, it is unlikely that a further decrease in $T_a$ will yield longer hibernation bouts in *E. telfairi*. The lesser hedgehog tenrec has a very poorly developed thermogenic capacity, possessing only very small amounts of brown adipose tissue and so relies largely on shivering for thermogenesis (Scholl, 1974). Together with the observation that maximal $T_b$ decreased with decreasing $T_a$, this indicates that there is a lower thermal limit from which *E. telfairi* cannot actively arouse. The two "unsuccessful" arousals observed during $W_{low}$ suggest that the lower limit of $T_a$ is about 12°C. Support for this comes from Scholl (1974), who noted that *E. telfairi* was not able to arouse successfully at a $T_a$ of 11°C. This sets an ultimate lower limit of $T_a$ for long-term survival in this species and restricts its potential habitats.

Besides the limitations of temperature-dependence of hibernation patterns on habitat choice in the tropical *E. telfairi*, such thermoregulatory plasticity would open adaptive advantages for opportunistic hibernators. As more field studies on hibernation are published, it is apparent that numerous tropical species use a flexible hibernation pattern, with some individuals within a population using hibernation, while others utilize daily torpor or stay euthermic (Kobbe and Dausmann, 2009; Lehmer et al., 2006; Schmid and Ganzhorn, 2009). This confirms that hibernation is not a fixed trait determined solely by physiological capacity but an adaptive behaviour controlled and modulated by individual and environmental factors.

It is therefore surprising that to date no field observations of inter-hibernation bout foraging trips have been made. This type of facultative hibernation (i.e. using hibernation bouts when environmental conditions are adverse and becoming active when they are good and when food/water availability is adequate) would be evolutionary and ecologically beneficial for hibernators, especially in unpredictable and changing environments. There would be several advantages to this approach. The most immediate is that it allows the animals to forage and replenish energy stores when food availability during the hibernation season is good, which will normally be when $T_a$ is high. This leads to greater energy stores on emergence after the hibernation season and thus more energy can be invested in reproduction. It might also be possible that due to higher energy stores, the animal could emerge earlier. Furthermore, facultative hibernators would not have to cease their territorial activities completely.

That inter-hibernation bout foraging is possible can be seen in hibernators that hoard food. Many rodents, e.g. members form the families Cricetidae, Gliridae and Sciuridae, support much of the energy needed for hibernation by hoarded food and consume the stored food during arousals (Vander Wall, 1990). Because food has to be digested and metabolized during the arousals, these euthermic phases are four times longer than that observed in fat-storing hibernators (Humphries et al., 2001). In fat-storing hibernators, which store the energy within their bodies, the digestive system often undergoes marked atrophy during hibernation (Carey, 1995), but digestive capability can be restored quickly after prolonged periods of fasting (Pennisi, 2005).

Therefore, why haven't facultative hibernators been found? It might be difficult for temperate and arctic hibernators to monitor and respond to favourable times during the hibernation season. Because most of these species hibernate in well-buffered burrows and caves (Buck and Barnes, 1999; Arnold et al., 1991) short-term changes in $T_a$ might not be detectable. Furthermore, arctic and temperate habitats have a more predictable seasonal cycle and thus might offer only transient opportunities for facultative hibernation. This in turn means that hibernators form these regions might be better off to continuously hibernate the whole winter.

The situation is different in the tropics. Although wet and dry season are predictable as well in most places, the differences between these seasons are not as pronounced as in arctic and temperate regions. Tropical hibernators within their often poorly insulated hibernacula (Dausmann et al., 2005; Schmid, 1998) could potentially sense changes in $T_a$, or even humidity, more easily and take advantage of more favourable conditions. Stephenson and Racey (1994) described the lowland streaked tenrec *Hemicentetes semispinosus,* a facultative hibernator that frequently arouses during the winter to forage. However, these authors made no continuous measurements of $T_b$ or $VO_2$ and did not present any details on pattern of hibernation or torpor. Nevertheless, this might be the first incidence of facultative hibernation. The closely related highland streaked tenrec *H. nigriceps* lives at higher altitudes and is thought to be an obligate hibernator. In contrast, the lowland streaked tenrec lives in a warmer habitat and remains active during favourable climatic conditions (Stephenson and Racey, 1994). This allows for winter breeding and a greater annual reproductive output in this species.

Staying inactive in a tree-hollow has been suggested to increase the probability of survival because of a lowered predation risk (Schmid and Kappeler, 1998). This might be one reason why more tropical mammals do not use an approach similar to that found in *H. semispinosus*. Alternatively it might be counterproductive for tropical species to become active at high $T_a$ if they use hibernation mainly for water conservations because higher $T_a$ will increase evaporative water loss. The mouse lemur *M. griseorufus* shows that at least one tropical hibernator has the right physiological capacities to potentially use facultative hibernation. Members of the same population of this species behave differently to cope with the unpredictability of their habitat (Kobbe et al., in manuscript). Some individuals use sporadic short torpor bouts, others daily torpor, short hibernation bouts or even long hibernation bouts. The seasonal accumulation of body fat partly determines which option is employed. Furthermore, it remains possible for individuals to vary their reliance on each of these strategies. *E. telfairi* might have similar behavioural options because it displays a variable hibernation pattern, even at the individual level, with the duration of hibernation/torpor bouts ranging from one day to over a week.

Another difference between tropical and arctic/temperate hibernation patterns is the duration of arousals. Re-warming from a hibernation bout is followed by a normothermic period of varying length before the animal resumes hibernation. Arousal duration in the ground squirrel *Spermophilus citellus* is about 15 to 18 h (Luis and Hudson, 2006; Hut et al., 2002) but much longer arousals are found in the European marmot *Marmota marmota* (Heldmaier et al., 2004). In marmots the re-warming period is followed by euthermic periods of one to two days. Long euthermic periods are required in food-hoarding hibernators to ingest and metabolize food and replenish energy stores (Humphries et al., 2001). Although arousal durations of more than 15 h seem to be the norm in temperate and arctic mammals, some small hibernators display shorter arousals, including the edible dormouse *G. glis*, in which arousals last for only 9 h (Wilz and Heldmaier, 2000). Duration of arousal seems to be affected by $T_a$, although not consistently. In ground squirrels of the species *Spermophilus beldingi* and *S. lateralis* longer arousals are found at higher $T_a$ (French, 1982), but higher $T_a$ leads to shorter arousals in the edible dormouse *G. glis* (Wilz and Heldmaier, 2000). The arousal duration of *E. telfairi* (11.8 h during the $W_{low}$ regime and 9.3 h under $W_{const}$) is clearly shorter compared to arctic and temperate hibernators, but nevertheless high compared to other tropical hibernators. The duration of arousal of the primate *C. medius* is less than 6 h (Dausmann et al., 2005) and can be as low as 3 h in the subtropical hibernating bat *Nyctophilus bifax* (Stawski et al., 2009). It is not clear why tropical hibernators exhibit shorter arousals. A general effect of $T_a$ is conceivable, especially since arousal duration is affected by $T_a$. However, the duration of arousals in the lesser hedgehog tenrec was not affected by $T_a$ and the bat *N. bifax* even increases arousal duration with increasing $T_a$ (Stawski et al., 2009).

### 4.3.2 Energy savings

Hibernation in temperate and arctic regions offers potentially enormous energy savings of up to 90% compared to the summer (Heldmaier et al., 2004). While MR during hibernation is on average reduced to 5-30% of BMR, it can be as little as 1% of RMR (Geiser and Ruf, 1995; Geiser, 2004) and energy savings of 98% compared to the euthermic state have been found in Richardson's ground squirrel *Spermophilus richardsonii* (Wang, 1978). However, these animals hibernate at a very low $T_a$ and show long hibernation bouts, and because most energy during hibernation is used during arousals (Boyer and Barnes, 1999), the energy savings are maximized.

In contrast tropical hibernators are characterized by opposite patterns of relatively high $T_a$ during hibernation and usually short hibernation bouts, at least at constant $T_a$. Nevertheless, tropical hibernation has the potential to realize significant energy savings as well as its beneficial effect on water usage (Cooper et al., 2005; Schmid and Speakman, 2000). Metabolic rate during hibernation bouts in the lesser hedgehog tenrec was reduced to 0.06 $ml_{O_2}g^{-1}h^{-1}$ during $W_{const}$, which is only 10% of normothermic MR. Even compared to MR during daily torpor bouts ($S_{const}$), this corresponds to a reduction of over 65%. This illustrates the energy-saving potential of tropical hibernation. Overall daily MR was reduced by 50% compared to the summer. Although this reduction did not reach statistical significance, it is potentially even larger since the summer $VO_2$ values are likely to be underestimates (see section 4.1.3 for full discussion). Another tropical hibernator from Madagascar, the fat-tailed dwarf lemur *C. medius* can lower its MR by 90% during hibernation bouts and reduces its energy requirements by 70% during hibernation (Dausmann et al., 2009). The observed MR reductions of *E. telfairi* are thus comparable to other tropical species.

The reason for the reduced energetic benefit of hibernation in the tropics compared to temperate and arctic regions is most likely the high $T_a$ at which tropical mammals hibernate. Temperate and arctic hibernators display a constant MR over a wide range of $T_a$ and thus $T_b$. This temperature independence during steady-state hibernation bouts has been found in marmots, eastern pygmy possums and ground squirrels (Arnold et al., 1991; Song et al., 1997; Buck and Barnes, 2000). Metabolic rate thus stays stable over a range of $T_a$. In contrast to BMR, there is no allometric relation between the MR during hibernation bout and body size. Metabolic rate during hibernation bouts appears to be a constant at 0.03 $ml_{O_2}g^{-1}h^{-1}$ for all hibernators (Heldmaier et al., 2004). If $T_a$ falls below the thermally optimal temperature range, MR increases because the animals actively regulate their $T_b$ which is now markedly above $T_a$ (Ortmann and Heldmaier, 2000; Buck and Barnes, 2000). The same happens when $T_a$ exceeds the optimal thermal zone (Ortmann and Heldmaier, 2000; Buck and Barnes, 2000), indicating that temperature independence of MR occurs only within a certain temperature range. However, some species do not display temperature independence of MR. The ground squirrel *S. lateralis* continuously increases its MR during hibernation when $T_a$ is increased and at 14°C consumes nearly three times as much oxygen as it does at 5°C (Zimmer and Milsom, 2001).

Metabolic rate during hibernation in tropical mammals is noticeably higher. The lesser hedgehog tenrec consumed 0.06 $ml_{O_2}g^{-1}h^{-1}$ during hibernation bouts ($W_{const}$), which is twice as much as temperate and arctic hibernators, whereas the minimum observed value of 0.03 $ml_{O_2}g^{-1}h^{-1}$ was similar to that in non-tropical species. The tropical dwarf lemur *C. medius* also consumes 0.06 $ml_{O_2}g^{-1}h^{-1}$ during hibernation bouts (Dausmann et al., 2009). This is an indication that tropical hibernators in general consume more energy during hibernation.

Whether tropical hibernation at low $T_a$ is more efficient than at high $T_a$ remains uncertain because there are no data on $VO_2$ for the $W_{low}$ regime. However, there is some indirect evidence on the subject. First, MR during hibernation is temperature-dependent. When animals were hibernating under the fluctuating $T_a$ regime, $VO_2$ correlated with $T_a$ and was lower during the colder night phase compared to the warmer day phase. However in a manner similar to that seen in arctic and temperate hibernators, $VO_2$ stayed constant over the range of $T_a$ from about 13°C to 20°C and increased if $T_a$ rose above that range of $T_a$ (see fig. 3.2.10). Further evidence comes from observations that $VO_2$ during hibernation bouts under the $W_{const}$ regime was markedly higher than $VO_2$ during daily torpor bouts under the $S_{const}$ regime. Together with the longer duration of hibernation bouts at lower $T_a$ and thus fewer, energetically-costly arousals, it is likely that it is energetically favourable for *E. telfairi* to hibernate at lower $T_a$.

### 4.3.3 Effects of fluctuating ambient temperature

One characteristic of tropical hibernation is that it occurs at high $T_a$. A second is frequency of significant fluctuations of daily $T_a$. Tropical hibernators can experience daily temperature fluctuations in their hibernaculum of over 20°C (Kobbe and Dausmann, 2009; Dausmann et al., 2005). The amplitude of these fluctuations is not only determined by changes in air temperature, but also by the insulating properties of the hibernaculum. Tree-hollows, which are often used as hibernacula by tropical mammals, display different insulating properties depending on the the size and species of tree (Dausmann et al., 2005; Schmid, 1998). The magnitude of changes in daily $T_a$ during hibernation is thus largely determined by the choice of hibernaculum. *E. telfairi* hibernates in tree-hollows, under logs on the ground or beneath the bark of a tree (Eisenberg and Gould, 1970). It seems to be quite opportunistic in its choice of hibernaculum and should therefore experience considerable fluctuations of $T_a$ in the wild.

Fluctuating $T_a$ can influence the pattern of hibernation, especially the occurrence of arousals. The factors triggering an arousal are not yet understood, but it has been suggested that $T_a$ triggers an arousal if it exceeds a critical value (Körtner and Geiser, 2000). This critical arousal temperature is not fixed for a given species, but can vary individually as well as through time (Turbill et al., 2008). A critical arousal temperature has been observed in a tropical hibernator, the mouse lemur *M. murinus* (Schmid, 1998). *M. murinus* can maintain torpor only below a threshold temperature of 28°C. If it is passively heated above this temperature, it actively arouses.

The existence of a critical arousal temperature for *E. telfairi* was not examined in the present study. Nevertheless, fluctuating $T_a$ had some effect on the occurrence of arousals. The average duration of hibernation bouts was reduced by over 50% under the $W_{fluc}$ regime compared to the $W_{const}$ regime and thus the frequency of arousals increased. This suggests that there might also be a critical arousal temperature for the lesser hedgehog tenrec. A daily maximum $T_a$ of 24°C, as used during $W_{fluc}$, might not have been sufficiently high to trigger an arousal, but the recurring high $T_a$ evidently did increase the propensity for arousals. An altogether different hibernation pattern is found in the dwarf lemur *C. medius* (Dausmann et al., 2005, 2004). Some individuals of that species hibernate in poorly insulated tree hollows and so experience large fluctuations in $T_a$. If this primate is passively heated to a $T_b$ of over 30°C on a regular basis, it continues to hibernate for weeks or even months without any arousals. If *C. medius* chooses tree hollows with good insulating properties, and thus experiences less pronounced $T_a$ fluctuations, it displays periodic arousals as do other hibernators. Whether a similar pattern could be found in the lesser hedgehog tenrec as well should be investigated in future studies.

As has been already described (section 4.2.3), re-warming costs can be largely reduced by passive heating (Schmid et al., 2000; Lovegrove et al., 1999b; Mzilikazi et al., 2002), which is especially beneficial because most energy during the hibernation season is consumed during arousals (Boyer and Barnes, 1999). Despite being well-known for its energetic benefit during daily torpor, the effect of fluctuating $T_a$ on arousals during hibernation has not been studied in detail. However, there are sources of external heat beyond that of ambient air or radiation. Young alpine marmots use the external heat provided by their parents to reduce the costs of re-warming (Arnold, 1993). Although this example demonstrates that passive warming is not limited to tropical hibernators, passive heating should occur most frequently in the tropics because of the high $T_a$ fluctuations found there.

Australian bats of the genus *Nyctophilus,* that hibernate in a mild climate, use the daily $T_a$ cycle to assist their arousals (Turbill and Geiser, 2008). They usually arouse after $T_b$ has been substantially raised by external heat. *E. telfairi* follows a similar pattern and arouses when $T_b$ has been increased passively by $T_a$. This is energetically advantageous and reduces arousal costs by nearly 70%. If we consider only the warming phase up to normothermic $T_b$, the energy savings amount to 85%, again highlighting the enormous benefit of passive heating. Further evidence of the energetic benefits comes from the observation of higher maximal $T_b$, that were attained during arousals. Although the difference did not reach statistical significance, maximal arousal $T_b$ was 1.5°C higher during the $W_{fluc}$ regime compared to the $W_{const}$ regime. This could be particularly advantageous in a species that has only a limited thermoregulatory capacity, as the lesser hedgehog tenrec.

In spite of the large effect of fluctuating $T_a$ on arousal costs, average daily $VO_2$ was not affected and did not differ when compared to that observed under the $W_{const}$ regime. Because hibernation bout $VO_2$ was the same for both winter conditions, the lack of effect of fluctuating $T_a$ on daily energy requirements can only be attributed to the higher frequency of arousals during $W_{fluc}$. Although both the frequency and cost of arousal are affected by fluctuating $T_a$, this has no effect on overall energy expenditure. A similar result was observed in a field study by

Dausmann et al. (2009), who compared the energetic costs of hibernation by the lemur *C. medius* in tree hollows that varied in their insulative properties. Although the amplitude of fluctuations in $T_a$ ranged from over 20°C to only 2°C, this had no effect on overall energy consumption. It can therefore be assumed that the choice of hibernaculum, i.e. well- versus poorly-insulated, is of no apparent energetic importance for *E. telfairi* as well.

# Chapter 5

# Summary

Energy in the form of food is a important resource for animals. This is especially true for mammals which use considerably more energy than ectotherms. Evolution has thus led to diverse adaptations that lower energy consumption. Mammals from temperate and arctic regions often use torpor and hibernation to save energy. Torpor and hibernation are physiological states that are characterized by a reduced body temperature and metabolic rate. Hibernation bouts can last from several days up to several weeks and are interrupted by short and energetically-costly arousals during which the animals warm until they reach normal body temperatures.

However, animals from the tropics can use torpor and hibernation as well, albeit at higher ambient temperatures. These higher ambient temperatures could affect energy consumption. Depending on the choice of hibernaculum, tropical hibernators are not only exposed to high ambient temperatures but also to significant daily fluctuations of ambient temperature. This in turn could affect the frequency as well as the energetic costs of arousals since fluctuating ambient temperatures offer the opportunity of passive heating to aid restoration of normal body temperature.

The effect of ambient temperature on hibernation by a tropical mammal, the lesser hedgehog tenrec (*Echinops telfairi*), was investigated. *E. telfairi* is a small insectivore weighing 140 g, that is endemic to Madagascar and superficially resembles a European hedgehog. From a total stock of 27 animals, a randomized sample of tenrecs was exposed to different ambient temperatures in a climate chamber. The experimental regimes included constant and fluctuating ambient temperatures both during the summer ($S_{const}$: 25°C and $S_{fluc}$: 20-30°C) and during the winter ($W_{const}$: 20°C, $W_{low}$: 15°C and $W_{fluc}$: 15-25°C). Energy consumption of the animals was measured by respirometry while the animals stayed within nestboxes in the climate chamber. Body temperature was measured with miniaturized temperature loggers (iButton), that were taped to the animals' abdomen.

*E. telfairi* can be considered an obligate heterotherm because all animals entered torpor or hibernated on every day, except for one female that remained normothermic for more than 24 h

on two occasions. Obligatory heterothermy and a lack of thermogenic capacity in this species are likely an ancestral trait. Body temperature in the summer followed ambient temperature closely during daily torpor bouts and reached maxima of 30.8°C ($S_{const}$) and 29.8°C ($S_{fluc}$) during the activity phase. Normothermic oxygen consumption was 0.67 $ml_{O_2}g^{-1}h^{-1}$ for $S_{const}$ and 0.43 $ml_{O_2}g^{-1}h^{-1}$ for $S_{fluc}$. During torpor bouts, oxygen consumption was reduced by around 80% to 0.18 $ml_{O_2}g^{-1}h^{-1}$ and 0.08 $ml_{O_2}g^{-1}h^{-1}$ for $S_{const}$ and $S_{fluc}$, respectively. Overall daily energy expenditure was 0.38 $ml_{O_2}g^{-1}h^{-1}$ for $S_{const}$ and 0.16 $ml_{O_2}g^{-1}h^{-1}$ for $S_{fluc}$. Re-warming under regimes of fluctuating ambient temperature was largely a passive process, reducing the cost of re-warming by 75%. This led to a reduction of 60% in overall energy requirements. Furthermore, fluctuating ambient temperature acted as a *zeitgeber* during daily torpor and triggered and/or synchronized the activity phases.

The lesser hedgehog tenrecs hibernated during winter and hibernation bout duration was negatively correlated with ambient temperature. The duration of hibernation bouts averaged 4.3 d under the $W_{const}$ regime and 8.5 d under $W_{low}$ with the highest observed bout lasting 12 d. In the fluctuating ambient temperature condition, the duration of hibernation bouts was reduced by 50% to 1.9 d. During hibernation, body temperature followed ambient temperature closely. Body temperature maxima during arousals were 28.1°C and 29.5°C under the $W_{const}$ and $W_{fluc}$ regimes, respectively. Maximum body temperature at the lowest ambient temperature (15°C) dropped to 19.6°C, partly due to unsuccessful arousals. This suggests the existence of a lower temperature limit below which *E. telfairi* cannot successfully arouse and thus suggests a limited thermoregulatory ability. Oxygen consumption during hibernation was reduced to 0.06 $ml_{O_2}g^{-1}h^{-1}$ and 0.10 $ml_{O_2}g^{-1}h^{-1}$ for $W_{const}$ and $W_{fluc}$, respectively. Although energy savings by hibernation in tropical species appears lower than in arctic and temperate hibernators, the overall energy requirements were still reduced by 50% compared to the summer. The reduced effectiveness of tropical hibernation is presumably caused by the higher ambient temperature at which tropical mammals hibernate. Warming costs during arousals in fluctuating ambient temperature were reduced by 85%. Although passive heating reduced the energetic cost of arousals, overall energy consumption was unaffected by fluctuating ambient temperatures because of the more frequent arousals.

# Chapter 6

# Zusammenfassung

Energie in der Form von Nahrung stellt für Tiere eine entscheidende Ressource zum Überleben dar. Dies gilt insbesondere für Säugetiere, welche im Vergleich zu ektothermen Tieren einen deutlich höheren Energieverbrauch haben. Im Laufe der Evolution sind diverse Anpassungen entstanden, die den Energieverbrauch senken. So nutzen Säugetieren aus temperaten und arktischen Regionen häufig Torpor und Winterschlaf, um Energie zu sparen. Dabei handelt es sich um physiologische Zustände, die durch eine Absenkung der Körpertemperatur und der Metabolismusrate gekennzeichnet sind. Die mehrtägigen oder -wöchigen Winterschlafphasen sind durch kurze, energetisch kostspielige Aufwachphasen unterbrochen, in denen die Tiere ihre normale Körpertemperatur erreichen.

Doch auch tropische Tiere nutzen Torpor und Winterschlaf, allerdings bei höheren Umgebungstemperaturen, welche sich auf den Energieverbrauch der Tiere auswirken können. Abhängig von der Wahl ihres Winterschlafquartiers sind tropische Winterschläfer nicht nur hohen, sondern täglich stark fluktuierenden Umgebungstemperaturen ausgesetzt. Dies kann sich nicht nur auf die Häufigkeit, sondern auch auf die energetischen Kosten der Aufwachphasen auswirken, da fluktuierende Umgebungstemperaturen die Möglichkeit zur passiven Erwärmung bieten.

Die Auswirkungen der Umgebungstemperatur auf den tropischen Winterschlaf wurden beim kleinen Igeltenrek (*Echinops telfairi*), als einem Modellorganismus für tropischen Winterschaf, untersucht. *E. telfairi* ist ein kleiner, 140 g schwerer Insektenfresser, der endemisch auf Madagaskar vorkommt und äußerlich einem Europäischen Igel gleicht. Aus einem Grundbestand von 27 Tieren wurde jeweils eine randomisierte Auswahl von Igeltenreks in einer Klimakammer verschiedenen Umgebungstemperaturen ausgesetzt. Die Versuchsbedingungen umfassten konstante und fluktuierende Umgebungstemperaturen sowohl im Sommer ($S_{const}$: 25°C und $S_{fluc}$: 20-30°C) als auch im Winter ($W_{const}$: 20°C, $W_{low}$: 15°C und $W_{fluc}$: 15-25°C). Dabei wurde der Energieverbrauch der Tiere respirometrisch als Sauerstoffverbrauch innerhalb ihrer Nistboxen erfasst. Die Körpertemperatur wurde mit miniaturisierten Temperaturloggern (iButton) aufgezeichnet, welche mit Klebeband auf dem Abdomen der Tiere befestigt wurden.

*E. telfairi* kann als obligat heterotherm bezeichnet werden. Alle Tiere gingen jeden Tag entweder in Torpor oder Winterschlaf, mit der Ausnahme eines Weibchens welches zweimal länger als 24 Stunden normotherm war. Die obligate Heterothermie und die fehlende thermogenetische Kapazität sind wahrscheinlich ursprüngliche Merkmale. Während des Torpors im Sommer folgte die Körpertemperatur der Umgebungstemperatur und erreichte Maxima von 30,8°C ($S_{const}$) und 29,8°C ($S_{fluc}$) während der Aktivitätsphase. Der normotherme Sauerstoffverbrauch lag bei 0,67 $ml_{O_2}g^{-1}h^{-1}$ für $S_{const}$ und 0,43 $ml_{O_2}g^{-1}h^{-1}$ für $S_{fluc}$. Während der Torporphasen wurde der Sauerstoffverbrauch um 80% auf 0,18 $ml_{O_2}g^{-1}h^{-1}$ und 0,08 $ml_{O_2}g^{-1}h^{-1}$ für $S_{const}$ beziehungsweise $S_{fluc}$ reduziert. Der Gesamtenergieverbrauch pro Tag lag 0,38 $ml_{O_2}g^{-1}h^{-1}$ ($S_{const}$) und 0,16 $ml_{O_2}g^{-1}h^{-1}$ ($S_{fluc}$). Das Aufwärmen während der Aktivitätsphasen war unter fluktuierenden Umgebungstemperaturen zum großen Teil ein passiver Prozess. Dies führte zu einer Reduzierung der Aufwärmkosten um 75% und erniedrigte den Gesamtenergieverbrauch um 60%. Des weiteren fungierte die fluktuierende Umgebungstemperatur als ein Zeitgeber während des Torpors und synchronisierte die Aktivitätsphasen.

Der kleine Igeltenrek hielt während der Wintermonate Winterschlaf, wobei die Dauer der Winterschlafphasen negativ mit der Umgebungstemperatur korrelierte. Im Durchschnitt dauerten die Winterschlafphasen 4,3 Tage für $W_{const}$ und 8,5 Tage für $W_{low}$, wobei die längste gemessene Winterschlafphase 12 Tage betrug. Im Versuchsansatz mit fluktuierenden Umgebungstemperaturen verringerte sich die Dauer der Winterschlafphasen um 50% auf 1,9 Tage. Während des Winterschlafes folgte die Körpertemperatur der Umgebungstemperatur und erreichte in den Aufwachphasen Maxima von 28,1°C und 29,5°C für $W_{const}$, bzw. $W_{fluc}$. Die maximale Körpertemperatur während des kältesten Versuchsansatzes (15°C) fiel auf 19.6°C, teilweise durch unerfolgreiche Aufwärmversuche. Dies legt ein unteres Temperaturlimit nahe, unterhalb dessen sich *E. telfairi* nicht mehr erfolgreich aufwärmen kann und welches für eine eingeschränkte Fähigkeit zur Thermoregulation spricht. Der Sauerstoffverbrauch während der Winterschlafphasen fiel auf 0,06 $ml_{O_2}g^{-1}h^{-1}$ für $W_{const}$, bzw. 0,10 $ml_{O_2}g^{-1}h^{-1}$ für $W_{fluc}$ ab. Obwohl tropische Winterschlaf im Vergleich zu temperatem oder arktischem Winterschlaf weniger effizient ist, wurde der Gesamtenergieverbrauch im Vergleich zum Sommer um 50% reduziert. Die verringerte Effektivität von tropischem Winterschlaf ist vermutlich durch die höheren Umgebungstemperaturen bedingt, unter denen tropische Säugetiere Winterschlaf halten. Die Kosten der Erwärmung während der Aufwachphasen wurden durch fluktuierende Temperaturen um 85% erniedrigt. Obwohl passives Aufwärmen die Energiekosten für die Aufwachphasen senkte, blieb der Gesamtenergieverbrauch durch das häufigere Auftreten von Aufwachphasen unter fluktuierenden Temperaturbedingungen unbeeinträchtigt.

# Acknowledgements

During the seemingly endless years that this thesis took to reach completion, a lot of people contributed in various ways to my work.

**Thank you very much indeed!**

Special thanks go to the following people:

Prof. Kathrin Dausmann, for supervising my PhD project(s), for scientific expertise and advice during the experiments and writing of this thesis.

Prof. Jörg Ganzhorn, for helpful advice and support throughout my time as a PhD student.

Prof. William Foley, for reviewing the thesis and checking the English (Australian) grammar and spelling.

Prof. Heinz Künzle, for donating the hedgehog tenrecs to us and for advice on keeping them.

Prof. Tassilo Feuerer, for his generosity to let us use his climate chambers for considerable time.

Dr. Bastian Tiemann, for veterinary advice and treatment.

Susanne Kobbe, for scientific and non-scientific conservations and advice, and for sharing an office with me.

All PhD and diploma students, for their help in the lab, their advice and comments and for keeping me company over the years.

My family, my girlfriend and my friends, for their love and support throughout my studies.

# Bibliography

Ali, J. R. and Huber, M. 2010. Mammalian biodiversity on madagascar controlled by ocean currents. *Nature*, 463(7281):653–6.

Andrews, M. 2007. Advances in molecular biology of hibernation in mammals. *Bioessays*, 29(5):431–440.

Arlettaz, R., Ruchet, C., Aeschimann, J., Brun, E., Genoud, M., and Vogel, P. 2000. Physiological traits affecting the distribution and wintering strategy of the bat tadarida teniotis. *Ecology*, 81(4):1004–1014.

Arnold, W. 1993. *Energetics of social hibernation*, pages 65–80. Life in the cold: ecological, physiological and molecular mechanisms. Westview, Boulder.

Arnold, W., Heldmaier, G., Ortmann, S., Pohl, H., Ruf, T., and Steinlechner, S. 1991. Ambient temperatures in hibernacula and their energetic consequences for alpine marmots marmota marmota. *J Therm Biol*, 16:223–226.

Asher, R. 1999. A morphological basis for assessing the phylogeny of the "Tenrecoidea" (Mammalia, lipotyphla). *Cladistics-the International Journal of the Willi Hennig Society*, 15(3):231–252.

Audet, D. and Thomas, D. 1996. Evaluation of the accuracy of body temperature measurement using external radio transmitters. *Canadian Journal of Zoology-Revue Canadienne De Zoologie*, 74(9):1778–1781.

Barclay, R., Kalcounis, M., Crampton, L., Stefan, C., Vonhof, M., Wilkinson, L., and Brigham, R. 1996. Can external radiotransmitters be used to assess body temperature and torpor in bats? *Journal of Mammalogy*, 77(4):1102–1106.

Barclay, R., Lausen, C., and Hollis, L. 2001. What's hot and what's not: defining torpor in free-ranging birds and mammals. *Canadian Journal of Zoology-Revue Canadienne De Zoologie*, 79(10):1885–1890.

Barnes, B. 1989. Freeze avoidance in a mammal - body temperatures below 0-Degrees-C in an arctic hibernator. *Science*, 244(4912):1593–1595.

Bartels, W., Law, B., and Geiser, F. 1998. Daily torpor and energetics in a tropical mammal, the northern blossom-bat macroglossus minimus (Megachiroptera). *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 168(3):233–239.

Beard, L. and Grigg, G. 2000. Reproduction in the short-beaked echidna, tachyglossus aculeatus: field observations at an elevated site in south-east queensland. *Proc Linn Soc N S W*, 122:89–99.

Bennett, A. F. and Ruben, J. A. 1979. Endothermy and activity in vertebrates. *Science*, 206(4419):649–54.

Boyer, B. and Barnes, B. 1999. Molecular and metabolic aspects of mammalian hibernation - expression of the hibernation phenotype results from the coordinated regulation of multiple physiological and molecular events during preparation for and entry into torpor. *Bioscience*, 49(9):713–724.

Buck, C. and Barnes, B. 1999. Temperatures of hibernacula and changes in body composition of arctic ground squirrels over winter. *J Mammal*, 80:1264–1276.

Buck, C. and Barnes, B. 2000. Effects of ambient temperature on metabolic rate, respiratory quotient, and torpor in an arctic hibernator. *American Journal of Physiology-Regulatory Integrative and Comparative Physiology*, 279(1):R255–R262.

Buffenstein, R. 1985. The effect of starvation, food restriction, and water deprivation on thermoregulation and average daily metabolic rates in gerbillus pusillus. *Physiological Zoology*, 58(3):320–328.

Buffenstein, R. and Salton, J. 2003. Field thermoregulatory profiles in tenrecs from the rainforest and dryforest of madagascar. *Integrative and Comparative Biology*, 43(6):1041–1041.

Butler, P., Green, J., Boyd, I., and Speakman, J. 2004. Measuring metabolic rate in the field: the pros and cons of the doubly labelled water and heart rate methods. *Funct. Ecol.*, 18(2):168–183.

Carey, H. 1995. Gut feelings about hibernation. *News Physiol Sci*, 10(2):55–61.

Carey, H., Andrews, M., and Martin, S. 2003. Mammalian hibernation: Cellular and molecular responses to depressed metabolism and low temperature. *Physiological Reviews*, 83(4):1153–1181.

Christian, N. and Geiser, F. 2007. To use or not to use torpor? activity and body temperature as predictors. *Naturwissenschaften*, 94(6):483–487.

Coburn, D. K. and Geiser, F. 1998. Seasonal changes in energetics and torpor patterns in the subtropical blossom-bat syconycteris australis (megachiroptera). *Oecologia*, 113(4):467–473.

Cooper, C., McAllan, B., and Geiser, F. 2005. Effect of torpor on the water economy of an arid-zone marsupial, the stripe-faced dunnart (Sminthopsis macroura). *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 175(5):323–328.

Cryan, P. M. and Wolf, B. O. 2003. Sex differences in the thermoregulation and evaporative water loss of a heterothermic bat, lasiurus cinereus, during its spring migration. *J Exp Biol*, 206(Pt 19):3381–90.

Daan, S., Barnes, B., and Strijkstra, A. 1991. Warming up for sleep - Ground-Squirrels sleep during arousals from hibernation. *Neuroscience Letters*, 128(2):265–268.

Dausmann, K. 2005. Measuring body temperature in the field - evaluation of external vs. implanted transmitters in a small mammal. *Journal of Thermal Biology*, 30(3):195–202.

Dausmann, K., Glos, J., Ganzhorn, J., and Heldmaier, G. 2004. Physiology: Hibernation in a tropical primate - even in the wound-down hibernating state, this lemur can warm up without waking up. *Nature*, 429(6994):825–826.

Dausmann, K., Glos, J., Ganzhorn, J., and Heldmaier, G. 2005. Hibernation in the tropics: lessons from a primate. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 175(3):147–155.

Dausmann, K., Glos, J., and Heldmaier, G. 2009. Energetics of tropical hibernation. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 179(3):345–357.

Deavers, D. R. and Musacchia, X. J. 1980. Water metabolism and renal function during hibernation and hypothermia. *Fed Proc*, 39(12):2969–73.

Douady, C., Catzeflis, F., Kao, D., Springer, M., and Stanhope, M. 2002. Molecular evidence for the monophyly of tenrecidae (mammalia) and the timing of the colonization of madagascar by malagasy tenrecs. *Molecular Phylogenetics and Evolution*, 22(3):357–363.

Eisenberg, J. and Gould, E. 1970. *The Tenrecs: A Study in Mammalian Behaviour and Evolution*, volume 27 of *Smithonian Contributions to Zoology*. Smithonian Institution Press, Washington.

Else, P. and Hulbert, A. 1981. Comparison of the mammal machine and the reptile machine - Energy-Production. *American Journal of Physiology*, 240(1):R3–R9.

Farmer 2000. Parental care: The key to understanding endothermy and other convergent features in birds and mammals. *Am Nat*, 155(3):326–334.

Fowler, P. A. and Racey, P. A. 1988. Overwintering strategies of the badger, meles meles, at 57 °n. *J Zool London*, 214(4):635–651.

Fox, J. 2005. The r commander: A basic-statistics graphical user interface to r. *Journal of Statistical Computing*, 14(9):1–42.

Francis, A. J. P. and Coleman, G. J. 1990. Ambient temperature cycles entrain the free-running circadian rhythms of the stripe-faced dunnart, sminthopsis macroura. *Journal of Comparative Physiology A: Neuroethology, Sensory, Neural, and Behavioral Physiology*, 167(3):357–362.

French, A. R. 1982. Effects of temperature on the duration of arousal episodes during hibernation. *J Appl Physiol*, 52(1):216–20.

French, A. R. 1985. Allometries of the durations of torpid and euthermic intervals during mammalian hibernation: a test of the theory of metabolic control of the timing of changes in body temperature. *J Comp Physiol B*, 156(1):13–9.

Garbutt, N. 2007. *Mammals of Madagascar.* Yale University Press, New Haven, London.

Geiser, F. 1988. Reduction of metabolism during hibernation and daily torpor in mammals and birds - temperature effect or physiological inhibition. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 158(1):25–37.

Geiser, F. 1994. Hibernation and daily torpor in marsupials - a review. *Aust. J. Zool.*, 42(1):1–16.

Geiser, F. 2004. Metabolic rate and body temperature reduction during hibernation and daily torpor. *Annual Review of Physiology*, 66:239–274.

Geiser, F. 2007. Yearlong hibernation in a marsupial mammal. *Naturwissenschaften*, 94(11):941–944.

Geiser, F. and Brigham, R. 2000. Torpor, thermal biology, and energetics in australian long-eared bats (Nyctophilus). *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 170(2):153–162.

Geiser, F. and Drury, R. 2003. Radiant heat affects thermoregulation and energy expenditure during rewarming from torpor. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 173(1):55–60.

Geiser, F. and Kenagy, G. 1988. Torpor duration in relation to temperature and metabolism in hibernating Ground-Squirrels. *Physiological Zoology*, 61(5):442–449.

Geiser, F. and Ruf, T. 1995. Hibernation versus daily torpor in mammals and birds - physiological variables and classification of torpor patterns. *Physiological Zoology*, 68(6):935–966.

Geiser, F. and Turbill, C. 2009. Hibernation and daily torpor minimize mammalian extinctions. *Naturwissenschaften*, 96(10):1235–40.

Gould, E. 1965. Evidence for echolocation in the tenrecidae of madagascar. *Proceedings of the American Philosophical Society*, 109(6):352–360.

Gould, E. and Eisenberg, J. 1966. Notes on biology of tenrecidae. *Journal of Mammalogy*, 47(4):660–&.

Grigg, G., Beard, L., and Augee, M. 2004. The evolution of endothermy and its diversity in mammals and birds. *Physiological and Biochemical Zoology*, 77(6):982–997.

Grigg, G. C., Beard, L. A., and Augee, M. L. 1989. Hibernation in a monotreme, the echidna (tachyglossus aculeatus). *Comp Biochem Physiol A Comp Physiol*, 92(4):609–12.

Guppy, M. and Withers, P. 1999. Metabolic depression in animals: physiological perspectives and biochemical generalizations. *Biol Rev Camb Philos Soc*, 74(1):1–40.

Hammel, H., Dawson, T., and Adams, R. 1968. Total calorimetric measurements on citellus lateralis in hibernation. *Physiological Zoology*, 41:341–357.

Heldmaier, G. 2003. *Vergleichende Tierphysiologie, vol. 2*. Springer Verlag, Heidelberg.

Heldmaier, G., Ortmann, S., and Elvert, R. 2004. Natural hypometabolism during hibernation and daily torpor in mammals. *Respir Physiol Neurobiol*, 141(3):317–29.

Heldmaier, G., Ortmann, S., and Körtner, G. 1993a. Energy requirement of hibernating alpine marmots. In Carey, C., Florant, G., Wunder, B., and Horwitz, B., editors, *Life in the cold: ecological, physiological and molecular mechanisms*, pages 175–183. Westview.

Heldmaier, G. and Ruf, T. 1992. Body-Temperature and Metabolic-Rate during natural hypothermia in endotherms. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 162(8):696–706.

Heldmaier, G., Steiger, R., and Ruf, T. 1993b. Suppression of metabolic rate in hibernation. In Carey, C., Florant, G., and Wunder, B., editors, *Life in the Cold*, pages 545–548. Westview.

Heldmaier, G., Steinlechner, S., Rafael, J., and Latteier, B. 1982. Photoperiod and ambient temperature as environmental cues for seasonal thermogenic adaptation in the djungarian hamster, phodopus sungorus. *Int J Biometeorol*, 26(4):339–45.

Heldmaier, G., Steinlechner, S., Ruf, T., Wiesinger, H., and Klingenspor, M. 1989. Photoperiod and thermoregulation in vertebrates: body temperature rhythms and thermogenic acclimation. *J Biol Rhythms*, 4(2):251–65.

Hemmingsen, A. 1960. Energy metabolism as related to body size and respiratory surfaces and its evolution. *Rep Steno Mem Hosp Nord Insulinab*, 9:1–110.

Hickey, M. and Fenton, M. 1996. Behavioural and thermoregulatory responses of female hoary bats, lasiurus cinereus (chiroptera: Vespertilionidae), to variations in prey availability. *Ecoscience*, 4:414–422.

Holloway, J. C. and Geiser, F. 1996. Reproductive status and torpor of the marsupial sminthopsis crassicaudata: Effect of photoperiod. *Journal of Thermal Biology*, 21(5-6):373–380.

Hudson, J. 1978. Shallow, daily torpor: a thermoregulatory adaption. In Wand, L. and Hudson, J., editors, *Strategies in Cold: Natrual Torpidity and Thermogenesis*, pages 67–108, London. Academic Press.

Hulbert, A. and Else, P. 2000. Mechanisms underlying the cost of living in animals. *Annual Review of Physiology*, 62:207–235.

Humphries, M., Thomas, D., Hall, C., Speakman, J., and Kramer, D. 2002. The energetics of autumn mast hoarding in eastern chipmunks. *Oecologia*, 133(1):30–37.

Humphries, M., Thomas, D., and Kramer, D. 2003. The role of energy availability in mammalian hibernation: A cost-benefit approach. *Physiological and Biochemical Zoology*, 76(2):165–179.

Humphries, M. M., Thomas, D. W., and Kramer, D. L. 2001. Torpor and digestion in food-storing hibernators. *Physiol Biochem Zool*, 74(2):283–92.

Hut, R., Barnes, B., and Daan, S. 2002. Body temperature patterns before, during, and after semi-natural hibernation in the european ground squirrel. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 172(1):47–58.

Jury, M. 2004. *The Climate of Madagascar*, chapter The Climate of Madagascar, pages 75–87. University of Chicago Press.

Kelm, D. and von Helversen, O. 2007. How to budget metabolic energy: torpor in a small neotropical mammal. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 177(6):667–677.

Kleiber, M. 1932. Body size and metabolism. *Hilgardia*, 6:315–353.

Kobbe, S. and Dausmann, K. H. 2009. Hibernation in malagasy mouse lemurs as a strategy to counter environmental challenge. *Naturwissenschaften*, 96(10):1221–7.

Kobbe, S., Ganzhorn, J., and Dausmann, K. Malagasy mouse lemurs show flexible hibernation behaviour as an adaptation to unpredictable environments.

Körtner, G. and Geiser, F. 2000. The temporal organization of daily torpor and hibernation: Circadian and circannual rhythms. *Chronobiology International*, 17(2):103–128.

Koteja, P. 2000. Energy assimilation, parental care and the evolution of endothermy. *Proc Biol Sci*, 267(1442):479–84.

Koteja, P. 2004. The evolution of concepts on the evolution of endothermy in birds and mammals. *Physiological and Biochemical Zoology*, 77(6):1043–1050.

Kozlowski, J. and Konarzewski, M. 2005. Functional ecology; west, brown and enquist's model of allometric scaling again: the same questions remain. 19(4):739–743.

Kozlwoski, J. and Weiner, J. 1997. Interspecific allometries are by-products of body size optimisation. *American Naturalist*, 149:352–380.

Larkin, J. E. and Heller, H. C. 1996. Temperature sensitivity of sleep homeostasis during hibernation in the golden-mantled ground squirrel. *Am J Physiol*, 270(4 Pt 2):R777–84.

Lehmer, E., Savage, L., Antolin, M., and Biggins, D. 2006. Extreme plasticity in thermoregulatory behaviors of free-ranging black-tailed prairie dogs. *Physiological and Biochemical Zoology*, 79(3):454–467.

Liow, L. H., Fortelius, M., Lintulaakso, K., Mannila, H., and Stenseth, N. C. 2009. Lower extinction risk in sleep-or-hide mammals. *Am Nat*, 173(2):264–72.

Lovegrove, B. 2003. The influence of climate on the basal metabolic rate of small mammals: a slow-fast metabolic continuum. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 173(2):87–112.

Lovegrove, B. and Genin, F. 2008. Torpor and hibernation in a basal placental mammal, the lesser hedgehog tenrec echinops telfairi. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 178(6):691–698.

Lovegrove, B., Raman, J., and Perrin, M. 2001. Heterothermy in elephant shrews, elephantulus spp. (Macroscelidea): daily torpor or hibernation? *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 171(1):1–10.

Lovegrove, B. G., Lawes, M. J., and Roxburgh, L. 1999a. Confirmation of pleisiomorphic daily torpor in mammals: the round-eared elephant shrew macroscelides proboscideus (macroscelidea). *J Comp Physiol B*, 169(7):453–60.

Lovegrove, G., Körtner, G., and Geiser, F. 1999b. The energetic cost of arousal from torpor in the marsupial sminthopsis macroura: benefits of summer ambient temperature cycles. *J Comp Physiol B*, 169(1):11–8.

Luis, A. and Hudson, P. 2006. Hibernation patterns in mammals: a role for bacterial growth? *Functional Ecology*, 20(3):471–477.

Lund, S. 2009. Verdauungseffizienz des kleinen madegassischen igeltenreks echinops telfairi im jahreszeitlichen verlauf. Master's thesis, Hamburg University.

Macmillen, R. E. 1965. Aestivation in the cactus mouse, peromyscus eremicus. *Comp Biochem Physiol*, 16(2):227–48.

MacPhee, R. and Novacek, M. 1993. *Definition and relationship of Lipotyphla*, pages 13–31. Mammalian Phylogeny: Placentals. Springer Verlag, New York.

McCall, R. A. 1997. Implications of recent geological investigations of the mozambique channel for the mammalian colonization of madagascar. *Proc Biol Sci*, 264(1382):663–5.

McMahon, T. 1973. Size and shape in biology. *Science*, 179(79):1201–4.

Miller, S. and Harley, J. 1996. *Zoology*. WCB, Dubuque.

Mzilikazi, N., Lovegrove, B., and Ribble, D. 2002. Exogenous passive heating during torpor arousal in free-ranging rock elephant shrews, elephantulus myurus. *Oecologia*, 133(3):307–314.

Nicoll, M. 1985. Responses to seychelles tropical forest seasons by a Litter-Foraging mammalian insectivore, Tenrec-Ecaudatus, native to madagascar. *Journal of Animal Ecology*, 54(1):71–88.

Nicoll, M. 1986. Diel variation in Body-Temperature in Tenrec-Ecaudatus during seasonal hypothermia. *Journal of Mammalogy*, 67(4):759–762.

Nicoll, M. 2001. *Tenrecs*. The Encyclopedia of Mammals. Andromeda Oxford Ltd., Oxfordshire.

Nikaido, M., Cao, Y., Okada, N., and Hasegawa, M. 2003. The phylogenetic relationships of insectivores with special reference to the lesser hedgehog tenrec as inferred from the complete sequence of their mitochondrial genome. *Genes & Genetic Systems*, 78(1):107–112.

Ortmann, S. and Heldmaier, G. 2000. Regulation of body temperature and energy requirements of hibernating alpine marmots (Marmota marmota). *American Journal of Physiology-Regulatory Integrative and Comparative Physiology*, 278(3):R698–R704.

Pennisi, E. 2005. The dynamic gut. *Science*, 307(5717):1896–9.

Popov, V. I. and Bocharova, L. S. 1992. Hibernation-induced structural changes in synaptic contacts between mossy fibres and hippocampal pyramidal neurons. *Neuroscience*, 48(1):53–62.

Popov, V. I., Bocharova, L. S., and Bragin, A. G. 1992. Repeated changes of dendritic morphology in the hippocampus of ground squirrels in the course of hibernation. *Neuroscience*, 48(1):45–51.

Poppitt, S., Speakman, J., and Racey, P. 1994. Energetics of reproduction in the lesser hedgehog tenrec, echinops telfairi (Martin). *Physiological Zoology*, 67(4):976–994.

Prendergast, B., Freeman, D., Zucker, I., and Nelson, R. 2002. Periodic arousal from hibernation is necessary for initiation of immune responses in ground squirrels. *American Journal of Physiology-Regulatory Integrative and Comparative Physiology*, 282(4):R1054–R1062.

R Core Development Team 2004. *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna.

Rabinowitz, P. D., Coffin, M. F., and Falvey, D. 1983. The separation of madagascar and africa. *Science*, 220(4592):67–69.

Racey, P. and Stephenson, P. 1996. Reproductive and energetic differentiation of the tenrecidae of madagascar. *Biogeography and Madagascar*, pages 307–319 588.

Ruf, T. and Arnold, W. 2008. Effects of polyunsaturated fatty acids on hibernation and torpor: a review and hypothesis. *American Journal of Physiology-Regulatory Integrative and Comparative Physiology*, 294(3):R1044–R1052.

Schmid, J. 1998. Tree holes used for resting by gray mouse lemurs (microcebus murinus) in madagascar: Insulation capacities and energetic consequences. *International Journal of Primatology*, 19(5):797–809.

Schmid, J. 2001. Daily torpor in free-ranging gray mouse lemurs (Microcebus murinus) in madagascar. *International Journal of Primatology*, 22(6):1021–1031.

Schmid, J. and Ganzhorn, J. U. 2009. Optional strategies for reduced metabolism in gray mouse lemurs. *Naturwissenschaften*, 96(6):737–41.

Schmid, J. and Kappeler, P. M. 1998. Fluctuating sexual dimorphism and differential hibernation by sex in a primate, the gray mouse lemur ( microcebus murinus ). *Behavioral Ecology and Sociobiology*, 43(2):125–132.

Schmid, J., Ruf, T., and Heldmaier, G. 2000. Metabolism and temperature regulation during daily torpor in the smallest primate, the pygmy mouse lemur (Microcebus myoxinus) in madagascar. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 170(1):59–68.

Schmid, J. and Speakman, J. R. 2000. Daily energy expenditure of the grey mouse lemur (microcebus murinus): a small primate that uses torpor. *J Comp Physiol B*, 170(8):633–41.

Scholl, P. 1974. Temperature regulation in small madagascar hedgehog, Echinops-Telfairi (Martin, 1838). *Journal of Comparative Physiology*, 89(2):175–195.

Snapp, B. D. and Heller, H. C. 1981. Suppression of metabolism during hibernation in ground squirrels (citellus lateralis). *Physiological Zoology*, 54(3):297–307.

Snyder, G. K. and Nestler, J. R. 1990. Relationships between body temperature, thermal conductance, q10 and energy metabolism during daily torpor and hibernation in rodents. *J Comp Physiol B*, 159(6):667–75.

Song, X., Körtner, G., and Geiser, F. 1997. Thermal relations of metabolic rate reduction in a hibernating marsupial. *Am J Physiol*, 273(6 Pt 2):R2097–104.

Stanhope, M., Madsen, O., Waddell, V., Cleven, G., de Jong, W., and Springer, M. 1998a. Highly congruent molecular support for a diverse superordinal clade of endemic african mammals. *Molecular Phylogenetics and Evolution*, 9(3):501–508.

Stanhope, M., Waddell, V., Madsen, O., de Jong, W., Hedges, S., Cleven, G., Kao, D., and Springer, M. 1998b. Molecular evidence for multiple origins of insectivora and for a new order of endemic african insectivore mammals. *Proceedings of the National Academy of Sciences of the United States of America*, 95(17):9967–9972.

Stawski, C., Turbill, C., and Geiser, F. 2009. Hibernation by a free-ranging subtropical bat (Nyctophilus bifax). *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 179(4):433–441.

Stephenson, P. 1994. Resting Metabolic-Rate and Body-Temperature in the aquatic tenrec Limnogale-Mergulus (Insectivora, tenrecidae). *Acta Theriologica*, 39(1):89–92.

Stephenson, P. and Racey, P. 1993a. Reproductive energetics of the tenrecidae (Mammalia, insectivora) .1. the Large-Eared tenrec, Geogale-Aurita. *Physiological Zoology*, 66(5):643–663.

Stephenson, P. and Racey, P. 1993b. Reproductive energetics of the tenrecidae (Mammalia, insectivora) .2. the Shrew-Tenrecs, microgale spp. *Physiological Zoology*, 66(5):664–685.

Stephenson, P. and Racey, P. 1994. Seasonal-Variation in resting Metabolic-Rate and Body-Temperature of streaked tenrecs, hemicentetes nigriceps and H-Semispinosus (Insectivora, tenrecidae). *Journal of Zoology*, 232(2):285–294.

Storey, K. and Storey, J. 1990. Metabolic rate depression and biochemical adaption in anaerobiosis, hibernation and estivation. *Q Rev Biol*, 65:145–174.

Storey, K. B. 1997. Metabolic regulation in mammalian hibernation: enzyme and protein adaptations. *Comp Biochem Physiol A Physiol*, 118(4):1115–24.

Strijkstra, A. and Daan, S. 1997. Sleep during arousal episodes as a function of prior torpor duration in hibernating european ground squirrels. *Journal of Sleep Research*, 6(1):36–43.

Turbill, C. and Geiser, F. 2008. Hibernation by tree-roosting bats. *J Comp Physiol B*, 178(5):597–605.

Turbill, C., Körtner, G., and Geiser, F. 2003a. Natural use of heterothermy by a small, tree-roosting bat during summer. *Physiol Biochem Zool*, 76(6):868–76.

Turbill, C., Körtner, G., and Geiser, F. 2008. Timing of the daily temperature cycle affects the critical arousal temperature and energy expenditure of lesser long-eared bats. *Journal of Experimental Biology*, 211(24):3871–3878.

Turbill, C., Law, B. S., and Geiser, F. 2003b. Summer torpor in a free-ranging bat from subtropical australia. *Journal of Thermal Biology*, 28(3):223–226.

Vander Wall, S. 1990. *Foodhoarding in Mammals.* University of ChicagoPress, Chicago.

Walsberg, G. and Hoffman, T. 2005. Direct calorimetry reveals large errors in respirometric estimates of energy expenditure. *Journal of Experimental Biology*, 208(6):1035–1043.

Wang, L. 1978. Energetics and field aspects of mammalian torpor: the richardsons ground squirrel. In Wang, L. and Hudson, J., editors, *Strategies in Cold: Natrual Torpidity and Thermogenesis*, pages 109–145. Academic Press.

Warnecke, L., Turner, J., and Geiser, F. 2008. Torpor and basking in a small arid zone marsupial. *Naturwissenschaften*, 95(1):73–78.

West, G. B., Brown, J. H., and Enquist, B. J. 1997. A general model for the origin of allometric scaling laws in biology. *Science*, 276(5309):122–6.

Wilz, M. and Heldmaier, G. 2000. Comparison of hibernation, estivation and daily torpor in the edible dormouse, glis glis. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology*, 170(7):511–521.

Woodley, R. and Buffenstein, R. 2002. Thermogenic changes with chronic cold exposure in the naked mole-rat (heterocephalus glaber). *Comp Biochem Physiol A Mol Integr Physiol*, 133(3):827–34.

Zimmer, M. and Milsom, W. 2001. Effects of changing ambient temperature on metabolic, heart, and ventilation rates during steady state hibernation in golden mantled ground squirrels (Spermophilus lateralis). *Physiological and Biochemical Zoology*, 74(5):714–723.