

Telemetric investigation of hibernation and post hibernal normothermy of European hamsters kept under semi-natural conditions

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Introduction

European or common hamsters belong to the rodent family Cricetidae and are closely related to the more popular Syrian or golden hamsters. The European hamster is widely distributed in the temperate Western Palaearctis between 44 and 59 °N and 5 - 95 °E. In

Central Europe its range reaches the western border with disjunct local occurrences in France (Strasbourg), the Netherlands and Germany. Until most recently, the European hamster was feared as a pest animal in agriculture. During the last decades, however, there was a serious decline in the population numbers of the European hamster, which forced the German authorities to place the old pest animal on the red list of threatened species. European hamsters are solitary living medium sized (200 - 500 g) food generalists living in burrows. They reproduce 2 - 4 times a year with a mean litter size of 6 and reach an age of 3 - 4 years (Niethammer 1982).

During wintertime, all homeothermic animals living in cold and temperate latitudes as well as in continental climates need more energy to keep their body temperature (T_b) around 38 °C. In addition to low ambient temperature (T_a), food shortage further increases the need for energy conservation. Depending on body size, there are various strategies to reduce energy loss. Big animals can reduce energy expenditure to 50 % by growing thick winter fur whereas small mammals can use this strategy only to a certain degree because of their relatively large body surface (Heldmaier 1990). Reduction of body mass prior to winter and episodic or periodical reduction of T_b are very effective strategies to reduce total energy expenditure during the hibernation season, e.g. to 77 % of basal in the golden hamster (McKee and Andrews 1992).

Like many animals of seasonal changing habitats, the European hamster shows seasonal rhythms in body mass, reproduction and the control of T_b . Many experiments conducted under constant ambient conditions proved this rhythmicity to be at least partially endogenous: seasonal rhythms continue even in the absence of external triggers. In nature, this endogenous seasonality is synchronised to the external seasons by time cues like photoperiod and T_a (Masson-Pévet et al. 1994).

Thermo-sensitive telemetry is a rather suitable method for continuous recordings of T_b in undisturbed animals. This paper describes an experimental set-up in semi-natural conditions and presents preliminary results on the time course of T_b during hibernation and during the normothermic period immediately after termination of heterothermy. In addition, our experimental set-up allowed recordings of the frequency and duration of times during which the animals left their hibernacula and were located above the ground (above ground activity, AGA). A third variable recorded was overall locomotor activity (LA).

Materials and Methods

The thermo-sensitive HF-transmitters used were developed by the Technical Workshop of the University of Konstanz. They were of cylindrical shape with maximum dimensions of 1.8 (diameter) by 4.2 cm (height) and a maximum weight of 18.6 g, corresponding to approximately 5 % of the body weight of a medium sized European hamster. Dimensions

and weight were mainly due to the relatively large batteries producing a 3 Volt output for 950 mAh. Transmitters were encoated into a mixture of bees wax and paraffin suitable for implantation into the abdominal cavity of the hamsters.

In each of two winters (1994/95 and 1995/96), 12 European hamsters (6 females and 6 males) were individually housed in wire mesh cages of 0.8 m (length) by 0.8 m (width) by 1.1 m (height). Cages were buried 0.6 m into the ground of an outdoor enclosure near the university campus and filled with soil in order to allow the animals to dig their own burrow inside the cage. Each cage was equipped with two spiral shaped antenna fixed into the PVC platforms at the bottom and the top of the cage.

All transmitters used the same carrier frequency of 14 MHz. They were screened off each other by the metal cages in case of the 12 implanted transmitters and by a small faraday cage built around the reference transmitter (#13) used for the measurement of T_a at ground level (Fig. 1). Transmitter #14 was buried in the soil at about the same depth as the bottom of the cages and was used as a reference for soil temperature. Inside each cage, the signals of the transmitter could be received either by the top or by the bottom antenna. In order to achieve simultaneous recordings and to minimise the distance between transmitter and receiver, every transmitter had its own single-super-heterodyne receiver placed on top of the cage. Temperature encoding pulse frequencies were converted into digital signals and passed on as opto-coupled signals. For the measurement of locomotor activity, field

strengths of pulses were passed on as analogue opto-coupled signals. The recording interval and sensitivity of the receivers were controlled by an IBM compatible personal computer (PC) containing a microcontroller card and running on a special developed MS-DOS-program that processed and stored the data. When the recording was interrupted for a specified time, a switch of the antenna was triggered by the microcontroller. Temperatures of all 14 transmitters were simultaneously measured as moving averages on 32 pulse frequencies within the recording interval (usually set to 5 min). In order to eliminate recording artefacts, the moving average procedure required 6 pulse frequencies with a specifiable amount of constancy (maximum deviation usually set to be less than 0.5 %). AGA was recorded as the percentage of an interval, during which signals were received by the top antenna. In addition, differences between the field strength of consecutive pulses were used to compute LA in a second moving average procedure. Incoming data could be viewed on the PC screen and were stored on the hard disk of the PC as well as on a floppy disk.

Periodicities of biological rhythms such as daily cycles as well as oestrous cycles were verified using the Chi^2 -periodogram (Sokolove and Bushell 1978).

Results

Due to technical problems and natural mortality, only a total of 13 complete and 5 partial hibernation patterns (HPs) was recorded. Figure 2 shows a complete recording of the HP of a female hamster. As in all hibernating species studied so far, hibernation of the European hamster is organised in discrete "bouts" of hypothermia (hibernation bouts, HBs), separated from each other by inter-bout normothermy (IBN). Twelve hamsters showed a regular pattern of 7 - 44 deep HBs with a decline of T_b near to soil temperature (minimum $T_b = 0.97\text{ }^{\circ}\text{C}$) and durations $> 24\text{ h}$ (maximum bout length = 165.2 h). Three other animals showed only a few deep HBs that were mixed with shallow HBs of shorter duration and separated by rather long periods of IBN. Finally, 3 hamsters did not hibernate: they showed no HBs at all. Hibernation started between October 2 and January 8 and ended between January 29 and April 9. Most hamsters stayed in their burrow for 4 - 39 days before the first decline of T_b below $30\text{ }^{\circ}\text{C}$ and remained inside their burrow for 0.5 - 22 days after the final rewarming in spring. However, 7 out of 13 hamsters interrupted hibernation for brief periods of times and appeared above ground. These animals spent a total of 0.43 - 401.74 h above ground. In animals showing a regular pattern of HBs with only short periods of IBN (Fig. 2), hibernal AGA was absent, whereas short episodes of AGA occurred in animals showing dispersed HPs within extended times of IBN. Even in those hamsters that did not show any HB, AGA was considerably reduced as compared to normothermic times. These animals stayed in their burrows for periods of up to 4 weeks without any AGA.

Of the total duration of the hibernation season about 1.6 - 80.2 % were spent in hypothermia (T_b below 30 °C) and 19.8 - 98.4 % in IBN. Mean T_b during hibernation ranged between 13.46 and 37.47 °C. LA was reduced to 15 % compared to the normothermic conditions before and after hibernation. The longest HBs were observed between end of December and end of March.

Each HB could be subdivided into a period of cooling-down (entry), a plateau phase with T_b levels around T_a , and a warming-up interval (arousal) (Fig. 3). During entry into hypothermia, T_b decreased quite slowly indicating a passive cooling along the temperature gradient. However, the occurrence of temporary rewarmings accompanied by peaks in LA indicate a regulated rather than a purely passive process. In the beginning of arousals from hypothermia, T_b increased first slowly to about 10 °C and then rather rapid until reaching normothermic T_b . The explosive increase in T_b was accompanied by the first occurrence of LA indicating the begin of thermogenesis through shivering of the animal. During the plateau phase of the HB, every extreme change in ambient temperature was followed by a similar change of T_b . As a result, T_b fluctuated within a range of 10 °C without leading to an interruption of the typical time course of a HB (Fig. 3). Time-lagged correlations between T_a and T_b were a rather common phenomenon observed within the plateau phase of HBs. Nevertheless, some hamsters showed spontaneous fluctuations in T_b during the plateau phase that were not caused by T_a . These low amplitude oscillations (less than 3 °C)

showed a considerable regularity, increased in amplitude and finally lead to termination of the HB during the rising phase of the last oscillation.

During posthibernal normothermy, a 24h-rhythm was clearly visible in T_b , AGA and LA (Fig. 4). In addition to daily cycles, the recordings also allowed detection of the oestrous cycle in female hamsters. During the posthibernal time from spring until summer 1996, a four-day rhythmicity in the daily maximum of T_b was observed in 3 out of 5 female European hamsters. Identification of the oestrous cycle in females was also possible by the analysis of mean daily percentage of AGA in 2 out of 5 animals and within a single female by analysing the daily sum of LA. It has to be evaluated whether the analysis of the oestrus cycle can be used to determine the onset and offset of the reproductive period during long-term recordings.

Discussion

Telemetric measurements of T_b in undisturbed animals over the entire hibernation season are fundamental to investigate the temporal organisation of hibernation and its relationship to environmental variables. The present study introduces first results on the time course of T_b during a long-term registration of hibernation in European hamsters kept under semi-natural conditions. The recordings demonstrated a variety of individual differences in the pattern of heterothermy confirming previous studies under laboratory

(Waßmer & Wollnik 1996) as well as semi-natural conditions (Wollnik and Schmidt 1995). The finding that some animals showed only dispersed HPs or no torpor at all proved that this is clearly not an artefact occurring only in laboratory conditions. However, the exact nature of this behaviour remains unclear. On one hand, it may be that the failure to hibernate indicates some sort of constrain either because of unsuitable external conditions or because of some physiological problem of the animal. On the other hand, it may be that there is a genetically fixed percentage of animals not entering deep hibernation in a facultative hibernator as the European hamster and these animals may have some advantage at least during not so harsh winters.

The present study revealed a number of interesting features of the HP of European hamsters. For example, due to the special set-up of the two antennas, additional information was obtained on the general activity of the animals as well as their appearance above ground. As far as we know, this is the first study of hibernating animals with simultaneous measurements of T_b and above ground activity. These measurements demonstrated that some animals leave their burrow during IBN. However, AGA is a rather rare event observed only in those animals with dispersed HPs or no torpor at all. Information on AGA could also be used to verify the hypothesis that males end their hibernation period earlier than females, but remain in their burrows in order to develop their testes before emergence in spring (Michener 1992). The data available so far for the

European hamster did not reveal any sex-related difference in the timing or amount of hypothermia.

Furthermore, the present study revealed interesting information on the time course of T_b during hypothermia. For example, the decrease of T_b during entry into hypothermia was occasionally interrupted by a brief increase of T_b and accompanied activity suggesting a regulatory increase in the metabolic rate in order to slow down the cooling process (e.g. Fig. 3). Similar results were found in a laboratory study investigating the effects of experimental cooling in active European hamsters. T_b often dropped in a stepwise manner indicating a similar stepwise decrease of the homeostatic setpoint (Malisch et al. 1975). Furthermore, in a previous laboratory study on European hamsters, we observed a stepwise decrease of T_b from the daily minimum of normothermic T_b to short and shallow HBs and finally to deep HBs in the beginning of the hibernation season indicating a gradual change of the thermoregulatory set point (Waßmer and Wollnik 1996). During the plateau phase of HBs, T_b was closely correlated with T_a (Fig. 3). However, T_b remained constant or increased when T_a decreased below a certain critical value. The present results thus confirm previous studies demonstrating that thermoregulation is not arrested during deep hypothermia (Heller and Collivier 1974, Heller et al. 1977). In contrast to the rather common phenomenon of a time-lagged correlation between T_a and T_b during deep hibernation, some animals showed spontaneous low amplitude oscillations in T_b that were

not caused by changes in T_a . Arousal from such HBs occurred during the rising phase of the final oscillation. A phenomenon like this was recently reported from a laboratory study on ground squirrels kept under constant darkness and temperature (Grahn et al. 1994) and was interpreted as an indicator of endogenous control on bout duration. The time-course of arousals from HBs could be separated into a slow rewarming up to approximately 10 °C, followed by an extreme warming rate that was accompanied by increasing LA (Fig. 3). These findings support the view, that initial heat production is generated through nonshivering processes of uncoupled respiration in brown fat mitochondria, followed by shivering thermogenesis of involuntary tonic or rhythmic contractions of muscles (ATP-hydrolysis) (Heldmaier et al. 1989).

Finally, the present recordings demonstrated not only daily rhythms in T_b , AGA and LA during posthibernal normothermy, but also oestrous correlated changes in the daily maximum of T_b and mean daily AGA rather similar to oestrous correlated changes of T_b previously reported for golden hamsters (Refinetti and Menaker 1992), marsupials (Rose and Jones 1996) and rats (Kent et al. 1991). The rather high levels of AGA, reflected also in T_b , are caused by the high level of oestrogen during the night of oestrous (Takahashi and Menaker 1980) and will most likely increase the chances of encounter with a potential mate.

In conclusion, the experimental set-up of our study revealed a number of fascinating aspects on the ecology and physiology of hibernation in the European hamster under semi-natural conditions. Supported by the DFG (Wo 354/2).

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Figure captions

Fig. 1: Schematic diagram showing the principle setup of the telemetry system used in the present study. For further explanations, see materials and methods.

Fig. 2: Complete recording of the hibernation period of a female hamster, showing a regular hibernation pattern in the time course of body temperature (upper panel, solid line). Minimal body temperature during hibernation bouts tracked spontaneous changes of ambient temperature (lower panel - dotted line). Above ground activity (black columns) was absent from 28 days before the first decline of body temperature below 30 °C until 2 days after the final rewarming at the end of the winter, indicating the times of immergence into and emergence from the burrow. The high percentage of above ground activity in the beginning of the recording was caused by burrow construction activity of the animal.

Fig. 3: Time course of body temperature (T_b , solid line), ambient temperature (T_a , dotted line) and general locomotor activity (black columns) during a single hibernation bout. Although this figure shows some rather typical characteristics of a hibernation bout, it is an atypical example in the sense, that this particular animal had its hibernaculum at ground level and was, therefore, more exposed to spontaneous fluctuation in T_a than animals hibernating

underground. However, time-lagged correlation of T_a and T_b did not only occur within the above ground recording shown here, but was a rather common phenomenon observed within the plateau phase of hibernation bouts.

Fig. 4: Typical recording of body temperature (upper panel, solid line - right axis), above ground activity (columns - left axis) and general locomotor activity (lower panel, Bezier line - also left axis) from the normothermic period following hibernation. Field strength difference was multiplied by 5 in order to achieve the same axis scale as above ground activity. White and black bars on top of the graph indicate the hours of astronomical day and night, respectively. Above ground activity was correlated with high levels of body temperature and locomotor activity. In all 3 parameters a stable bimodal pattern was observed with one peak around 20H00 and a second peak around 04H00 corresponding to the hours of twilight.







