A C T A T H E R I O L O G I C A

VOL. XV, 24: 373—380. BIAŁOWIEŻA 15.XII.1970

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Initial Stages of Hibernation: Is Cold Acclimatization Necessary

[With 4 Figs.]

Four species of hibernators were exposed to a warm and then a cold environment to determine whether cold acclimatization is a prerequisite to dormancy. The four species tested in the cold in October were: hedgehogs, woodchucks, edible dormice and garden dormice. Hedgehogs, woodchucks and garden dormice are capable of hibernation within 48-hours after removal from a warm environment; individuals which hibernated directly after being placed in a cold environment had not shown thermal lability in the warm environment. Edible dormice seemed to require at least 10 days of cold preconditioning before entering hibernation. We concluded that cold acclimatization is not necessarily a prerequisite for dormancy. One woodchuck, monitored in detail, initiated all bouts of hibernation during the usual time of sleep. This woodchuck was monitored a second winter while hibernating outdoors; its hibernating heart rates fluctuated inversly with air temperature, showing a homeostatic relationship. Therefore, outdoor hibernation with changing air temperature is a more variable process than in the constant-temperature coldroom.

I. INTRODUCTION

When mammals are first exposed to continuous cold, physiological mechanisms for combating cold gradually change over at least a 10 day period (F o l k, 1966). The basal metabolic rate increases, the maximum metabolic rate rises, non-shivering thermogenesis increases, shivering decreases, and there is hypertrophy of endocrine and visceral organs. The present study concerns whether these physiological changes (called »cold-acclimatization«) are prerequisite to mammalian dormancy.

II. MATERIALS AND METHODS

Four species of hibernators were tested in October: three female hedgehogs, *Erinaceus europaeus* L i n n a e u s, 1758; three female woodchucks, *Marmota monax* (L i n n a e u s, 1758); four edible dormice (two female), *Glis glis* (L i n n a e u s, 1766); and four garden dormice (two female), *Eliomys quercinus* (L i n n a e u s, 1766). The hedgehogs were captured near Nijmegen. The Netherlands, and were maintained in captivity for two months before study in Nijmegen. The woodchucks were captured as young animals in Iowa, U.S.A., and were raised and studied at the University of Iowa. The dormice were purchased from a French animal supplier, who trapped them in France and shipped them to Nijmegen. They were maintained in the Nijmegen animal colony for one month before the study began.

The coldroom (hibernation) phase of the study was terminated after four weeks except with one monax which was studied for two winters. Two of the hedgehogs and two of the woodchucks did not hibernate within the first three weeks. Later it was learned that if food and water had been removed, it might have induced these animals to hibernation. All dormice did hibernate within the one month cold room period. The following physiological conditions were monitored with the rodents in a warm room, and then a coldroom ($5 \pm 1°C$) without restraint: skin temperature by recording-radiometer, body temperature and heart rate-*ECG* by the Iowa implantable radio-capsule (Folk, 1964). These radio-capsules are surgically inserted into the abdominal cavity. One of the woodchucks which did hibernate contained a 32 gram radio-capsule which gave good physiological data for two years with one winter in a coldroom and the second in a large outdoor cage. The outdoor cage contained abundant insulation, soil, and shredded newspaper and was covered with heavy canvas.

The studies on the hedgehog and the woodchuck included heart rate and *ECG* measurements. In addition, the woodchuck's body temperature was monitored during the second winter by means of radio-frequency detuning (see Results). Heart rates were continuously monitored by automatic recording for 30 sec each half hour (Folk, 1964).

Only the body temperatures of the dormice were monitored. This was done either by recording-radiometer or in the case of one *Glis* and one *Eliomys*, by implanted radio-capsule.

III. RESULTS

Three species entered hibernation 24—48 hours after being moved from the warm environment. The edible dormice, however, seemed to require at least 10 days of cold preconditioning.

Hedgehog. The hedgehog which hibernated had been maintained for three months after implantation of the transmitter. Its weight gain and behavior indicated good health. During the control period, just before exposure to cold in October (Fig. 1), sleeping and active heart rates were almost identical to those recorded from August 6 to cold exposure. The ten highest heart rates during the summer ranged from 273 to 320 b/m, usually between 08.00 hrs and 10.00 hrs. The ten lowest heart rates, 120 to 136 b/m usually occurred during sleep. Since these summer heart rates persisted to the time of cold-exposure, there is no indication that the hedgehog was experiencing either preliminary or partial lethargy, *i.e.*, it did not lower its body temperature toward room temperature ($26.5 \pm 1°C$, *RH* 60%). Once in the coldroom, the question was: does the animal need 10 days or more to cold-acclimatize before entering hibernation, or can he go right into hibernation in a day or two? The animal was not automatically recorded the first three days in the coldroom, but readings by stop-watch indicated that the heart rate was still very high after twenty hours. By the 46th hour he was in deep hibernation, heart rate 9—13 b/m; this is typical hedgehog hibernation as described by Kristoffersson & Soivio (1964). The first few bouts of hibernation were short which is typical of all mammalian hibernators (Folk, 1966). They then lengthened to 10—14 day bouts for the remainder of the experiment.

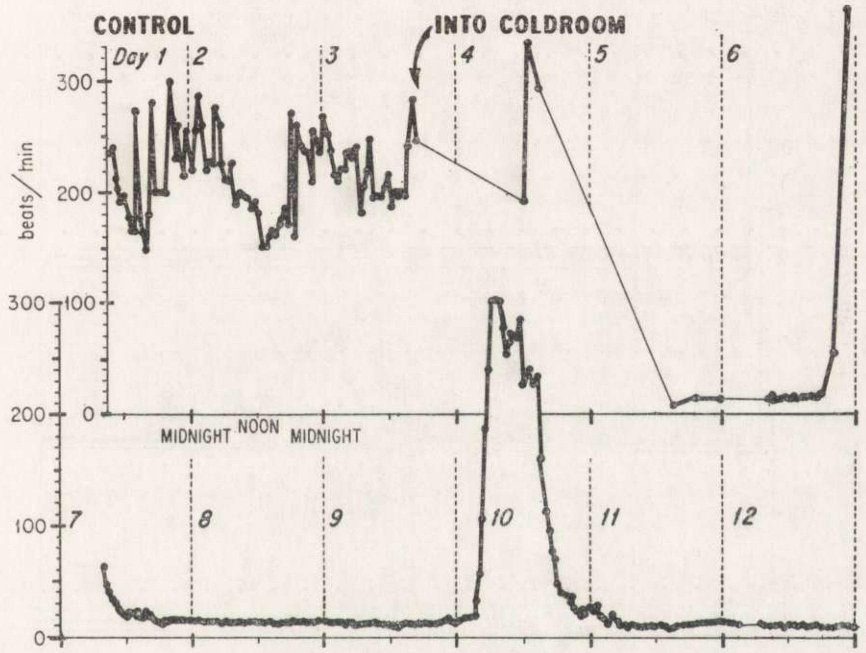

Fig. 1. Heart rates of non cold-acclimatized hedgehog entering first hibernation in a controlled coldroom (5 ± 1°C).

Woodchuck. The first year, first-hibernation was delayed for four weeks in the coldroom until the food and water were removed. After this it showed typical bouts of hibernation. Four bouts of awakening are compared in Fig. 2; it shows the duration of wakefulness and the abrupt increase in metabolic rate accompanied by an extremely rapid change in heart rate. The gradual return to the condition of dormancy is evident. The level of first-winter hibernating heart rates may be compared with those of its second winter, outdoors (Figs. 3 and 4). During the preceding summers, sleeping heart rates were 80 to 90 b/m. During the fall this rate lowered to 50 b/m. The animal had gained a great deal of weight in the warmroom, and undoubtedly the short natural photoperiod prepared it for hibernation, but this preparation cannot be called »cold-acclimatization«. This lowering of control sleeping rate had not occurred with the hedgehog.

When the woodchuck was moved to its cold outdoor enclosure, after 48-hr its heart rate dropped to 10 b/m and it was in deep hibernation. Eight hours later it was back to the normothermic state; then there was one hibernation-bout per day for three days. These short initial bouts

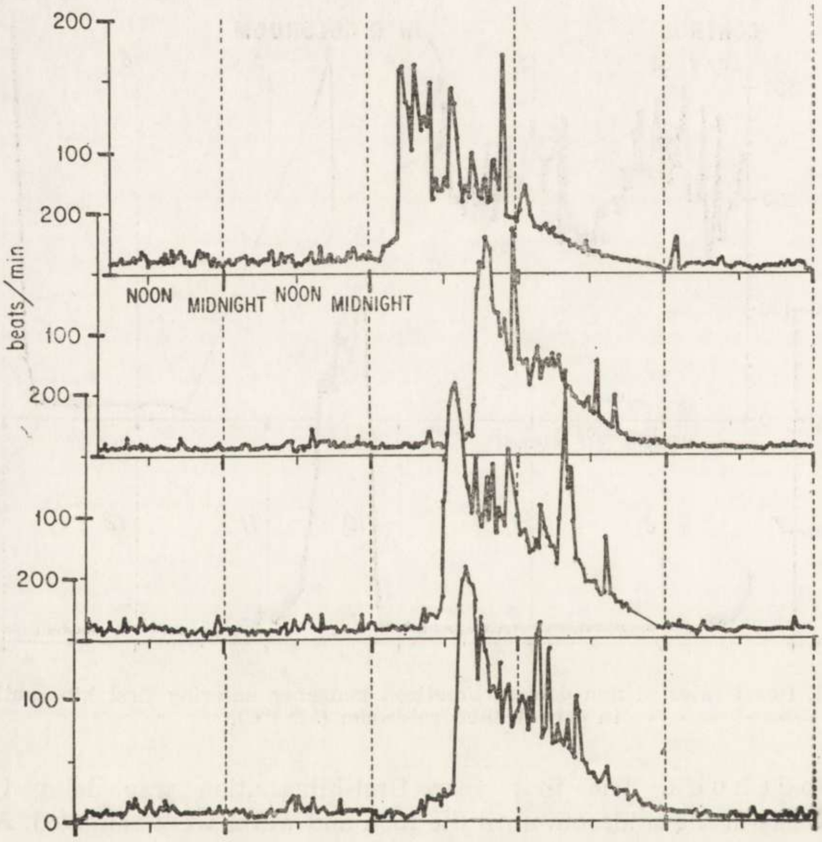

Fig. 2. Heart rates of woodchuck awakening from four bouts of hibernation in controlled coldroom (5±1°C) (first winter).

have been called »test drops«. Again, »cold-acclimatization« could not take place in this short time. Others have observed test drops, but ours are shorter; Strumwasser (1959) describes them in body temperatures that last two days per drop and 6 days to enter hibernation. However, his animals were cold-acclimatized before being tested. Our data here are unusual because the animal was monitored in the outdoor winter environment under variable air conditions, and yet qualitatively the animal displayed typical hibernation of the sort which was displayed in the coldroom the winter before. Quantitatively, the pattern is probably more typical of animals in the free environment; a remarkable illustration of homeostasis was evident as the hibernating woodchuck responded

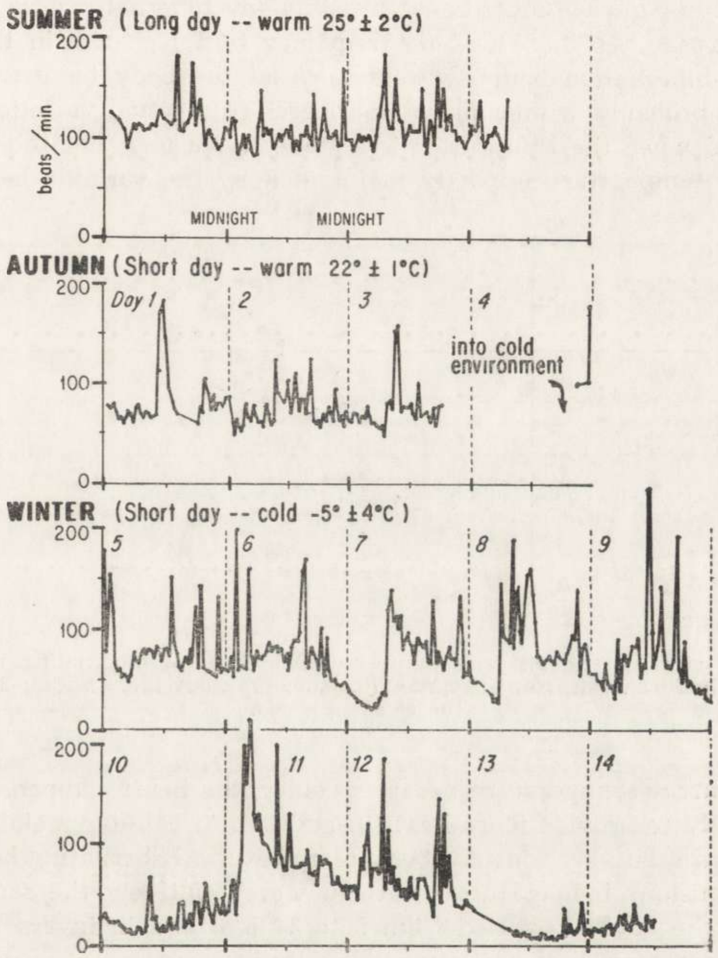

Fig. 3. Heart rates of non cold-acclimatized woodchuck entering first hibernation in the outdoor environment. Control heart rates for summer and autumn are presented in lines 1 and 2.

to an ambient temperature range of 50 degrees. Air temperatures around the burrow were monitored; however, we were unable to monitor temperatures of the inner-burrow or of the skin or fur of the animal. We have plotted, in Fig. 4, a selected series of readings consisting of only the hibernating heart rates at 08.00 hrs, and only those which occurred

24 hours before or after an awake period. During this steady state of hibernation, the body temperature showed little variation; probably no more than one degree. As our *ECG* transmitters are temperature sensitive, we have found that the receivers detune if there is a change in transmitter temperature (caused by a change in abdominal temperature). The receivers always must be adjusted as any hibernator goes in or out of hibernation. As there was no frequency (*KHz*) change in the course of these hibernation bouts, we assume that the body temperature was constant, probably within about one degree (the actual variation can be calculated when the transmitter is removed). We feel that the hibernating body temperature is partly maintained by the variable heart rates.

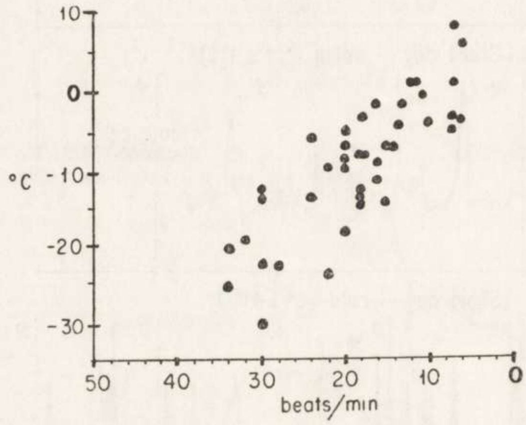

Fig. 4. Heart rates and air temperatures over burrow of hibernating woodchuck. The 08.00 hr heart rates for all winter are shown except those within 24 hours of the awake periods.

As the burrow temperature becomes colder, the heart compensates, perhaps partly to provide more heat (in addition to cellular metabolism) to maintain a relatively constant body temperature. Hibernating heart rates in the constant temperature coldroom were relatively the same, while those in the outdoors varied from 6 to 34 b/m and in inverse direction from the ambient.

Edible dormouse. Four specimens were studied by means of implanted temperature-capsules and by continous radiometer recordings. Skin temperatures in the warm control environment varied from 25.2 to 30.2°C in a day-night rhythm during October. A daily photoperiod of 8 hrs may have acted as a clue to seasonal adjustment. Nevertheless, these dormice, when moved in their own cages into the coldroom (*DD*), did not hibernate for at least 10 days; within four weeks all specimens hibernated.

G a r d e n d o r m o u s e. During October there were no important day-night variations in skin temperature, 29.7 to 31.6°C. This species, however, went into deep hibernation in the coldroom within 48 hours, except for one which required 7 days. During hibernation their skin temperatures went below 10°C; the radiometer pen went off-scale at 10°, but deep hibernation was apparent by visual monitoring of respiration. Lengths of hibernation-bouts were not obtained.

IV. DISCUSSION

These results indicate that cold-acclimatization is not necessarily a prerequisite for mammalian hibernation. The woodchuck, hedgehog and garden dormice were capable of hibernating two days after coldroom exposure with no cold preconditioning. We have found this situation in other species, but we had not monitored their warmroom condition (*i.e.*, were they already in a semi-dormant state in the warmroom?). In the present species, we established that the hedgehog, woodchuck and four garden dormice maintained normothermic conditions in the warmroom during the month of September. However, the edible dormice showed a wide day-night rhythm of body temperature, and a lack of coordination during the low phase which probably should be interpreted as daily semi-torpor. This condition did not appear to »prepare« them for hibernation.

Other species which are known to be capable of quickly entering hibernation are the 13-lined ground squirrel (F o l k, 1966; D a w e, 1970) and the Arctic ground squirrel (F o l k, 1967). However, S t r u m w a s s e r's (1959) California ground squirrels were pre-cold-exposed and they required six days to reach deep hibernation using test drops. The pattern in Fig. 3 seems to indicate woodchuck test-drops, but the process required only three days and there was no preparitory cold exposure.

The relationship of sleep period to hibernation entry time is interesting. The woodchuck entered hibernation during the natural sleep period both in the initial short bouts (Fig. 3) and in each successive bout (Fig. 2). This relationship deserves further analysis; perhaps knowledge of the sleep condition could assist us in understanding the nature of the hibernation mechanism (S o u t h, 1969). The results presented here also add to our understanding of the hibernation mechanism: while cold air is necessary for body temperature depression, previous appreciable exposure to cold air is apparently not necessary to prepare the hypothalmus, thyroid, adrenal glands, brain and heart so that they may permit deep hibernation.

Acknowledgement: Part of this work was accomplished while supported by a fellowship from the Fulbright Program of the United States Educational Foundation, held while in the Department of Physiology of the University of Nijmegen.

The generous assistance of Mr. J. N. Van Gaalen is gratefully acknowledged. Research support was provided by the Arctic Institute of North America under contract with the Office of Naval Research.

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Received, August 8, 1970.

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POCZĄTKOWE OKRESY HIBERNACJI: CZY JEST KONIECZNA AKLIMATYZACJA DO CHŁODU?

Streszczenie

Odpowiedzi na pytanie zawarte w tytule szukano na drodze eksperymentalnej. Zbadano 4 gatunki ssaków — jeża (*Erinaceus europaeus*), świstaka amerykańskiego (*Marmota monax*), popielicę (*Glis glis*) i żołędnicę (*Eliomys quercinus*). Jeż, świstak i żołędnica są zdolne do hibernacji w ciągu 48 godzin po przeniesieniu ich z ciepłego pomieszczenia do chłodnego. Osobniki, które zapadły w sen bezpośrednio po ich umieszczeniu w pomieszczeniu o niskiej temperaturze, nie wykazały labilności termicznej w wyższej temperaturze. Wydaje się, że popielica potrzebuje co najmniej 10 dni na aklimatyzację do chłodu, zanim rozpocznie hibernację. Sądzić jednak należy, że aklimatyzacja do warunków chłodu nie jest potrzebna przed okresem hibernacji. Jeden ze świstaków, badany szczegółowo, wykazywał objawy hibernacji w porze normalnego snu. Ten sam osobnik w ciągu drugiej zimy, spędzonej w warunkach naturalnych, wykazywał homestatyczną zależność, gdyż uderzenia serca w czasie snu zmieniały się odwrotnie niż temperatura powietrza. Wskaźniki fizjologiczne w czasie hibernacji w warunkach naturalnych, przy zmieniającej się temperaturze powietrza są bardziej zmienne, niż w czasie badań w warunkach laboratoryjnych.