mals (intact or deafferented cats) was carried out by analysis of variance (*8*); the results appear as probability in the last column of Table 1.

With intact animals, light sleep was accompanied by slight but definite decrease in both systolic and diastolic blood pressures (Table 1, Light sleep lowest). Pressure falls were more consistent during episodes of deep sleep. Although these falls were somewhat emphasized by a small rise in blood pressure at the very beginning of a deep-sleep period (Deep sleep initial), they were never dramatic (averaging 24.8 mm-Hg for systolic, and 18.6 mm-Hg for diastolic pressure), and there were no exceedingly low values of arterial pressure during the deep sleep of intact animals (Deep sleep lowest).

Changes in similar directions, but with some striking quantitative differences, were observed in animals with sino-aortic deafferentation. As indicated in Table 1, systolic and diastolic blood pressures were significantly higher in these than in intact animals both during quiet wakefulness (Waking) and throughout light sleep (Light sleep lowest), although under the latter condition a consistent decrease in pressure usually occurred, as previously reported for the dog (*9*). In spite of the fact that higher pressures were still observed in deafferented animals at the beginning of deep sleep (Deep sleep initial), such surprisingly large falls in pressure were recorded during the course of deep-sleep episodes (average falls: 59.4 and 47.5 mm-Hg for systolic and diastolic blood pressure, respectively) that arterial pressure finally attained much lower absolute values in deafferented than in intact animals (Deep sleep lowest). Not infrequently, such values were low enough to endanger an animal's survival. Systolic values were lower than 65 mm-Hg during deep sleep in 46 out of 145 episodes in deafferented animals, but in only 6 of 82 observed episodes in intact animals; likewise, diastolic values lower than 30 mm-Hg (never observed in intact animals) occurred in 63 of 145 episodes of deep sleep in deafferented cats. In several incidents of the lowest values of blood pressure, and particularly whenever diastolic pressure approached or attained 0 mm-Hg, the electrocorticogram rapidly flattened, and some convulsive waves heralded a generalized seizure (Fig. 1); all of a sudden the arterial pressure showed a prolonged hypertensive rebound and the animal awoke in fright, the electroencephalogram promptly returning to its normal waking appearance. These dramatic signs of transient cerebral ischemia, though observed in only a few deafferented animals, never occurred either in our groups of intact animals or in several hundred normal cats whose sleep behavior was recently studied in our laboratory.

Since section of the carotid sinus and aortic nerves interrupts both presso- and chemoceptive fibers, we cannot conclude that abolition of the pressoceptive rather than the chemoceptive reflexes is responsible for the falls in systolic and diastolic blood pressure to such dramatically low levels. It is difficult to explain the occurrence of lower pressures in deafferented than in intact animals, either with the classical conception that pressoceptive inactivation releases pressor centers from a tonic inhibitory inflow, or with the current opinion that the role of chemoceptors in circulatory regulation is limited to such emergencies as acute anemia or anoxia. While other explanations cannot be ruled out at present, our observations suggest that the actual functions of sino-aortic reflexes in unanesthetized normal animals may differ, at least in part, from the patterns inferred from the classical experiments on anesthetized or decerebrate animals.

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References and Notes

1. N. Kleitman, *Sleep and Wakefulness* (Univ. of Chicago Press, Chicago, 1963), pp. 42–47.
2. O. Candia, E. Favale, A. Giussani, G. F. Rossi, *Arch. Ital. Biol.* **100**, 216 (1962); E. Kanzow, D. Krause, H. Kunhel, *Arch. Ges. Physiol.* **274**, 593 (1962); F. Bonamini, V. DeCarolis, P. Pastorino, G. F. Rossi, *Riv. Neurobiol.* **8**, 394 (1962).
3. F. Snyder, J. A. Hobson, D. F. Morrison, F. Goldfrank, *J. Appl. Physiol.* **19**, 417 (1964).
4. G. Moruzzi, *Harvey Lectures* **58**, 233 (1963).
5. C. Heymans and E. Neil, *Reflexogenic Areas of the Cardiovascular System* (Churchill, London, 1958); M. Guazzi, A. Libretti, A. Zanchetti, *Circulation Res.* **11**, 8 (1962).
6. Sponsored by the European office of the U.S. Air Force Office of Scientific Research, OAR (grant AF EOAR 64-41), and by Consiglio Nazionale delle Ricerche (Electrophysiology Group).
7. Battaglia-Rangoni M. 10.
8. G. W. Snedecor, *Statistical Methods* (Iowa State College Press, Ames, 1946).
9. A. Samaan, *Compt. Rend. Soc. Biol.* **115**, 1383 (1934); C. Heymans and J. J. Bouckaert, *ibid.* **117**, 252 (1934).

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Impaired Recovery from Hypothermia after Anterior Hypothalamic Lesions in Hibernators

Abstract. *Hypothermia was induced by hypercapnic hypoxia in thirteen-lined ground squirrels,* Citellus tridecemlineatus. *When the squirrels were allowed to recover normal body temperatures at a 10°C ambient temperature, those with anterior hypothalamicpreoptic lesions took three to four times longer than normal controls to reach a body temperature of 35°C.*

The anterior hypothalamic-preoptic area plays an important role in physiological (*1*) and behavioral (*2*) temperature regulation in homeothermic animals. Animals with lesions in this area lose the ability to maintain normal temperatures when exposed to a cold environment (*3*). Nothing is known, however, of the function of this region in hibernators. The experiment described here concerns the effects of ablation of the rostral hypothalamus on recovery from induced hypothermia in the thirteen-lined ground squirrel, *Citellus tridecemlineatus.*

Twenty-four female squirrels weighing from 190 to 300 g were used. Sixteen were operated on, each being given a standard dose of 0.4 ml atropine sulfate (U.S.P. grade) (concentration 0.4 mg/ml) before being anesthetized with Nembutal (50 mg/kg of body weight) injected intraperitoneally. The scalp was incised, and a hole drilled stereotaxically over the superior sagittal sinus. Bilateral electrolytic lesions of varying sizes were made in the vicinity of the anterior hypothalamic-preoptic area. The stereotaxic coordinates found to be most effective in producing a deficit, with the animal's head in a horizontal position, were A 8.5, L 0.6, and 8.0 to 8.5 mm below the dural surface. The squirrels were operated on in pairs matched for weight, and after surgery each animal was given a standard intramuscular injection of 0.3 ml procaine penicillin (300,000 U/ml) and was put in an individual cage. At this time a control animal for each pair, matched as closely as possible for weight, was also taken

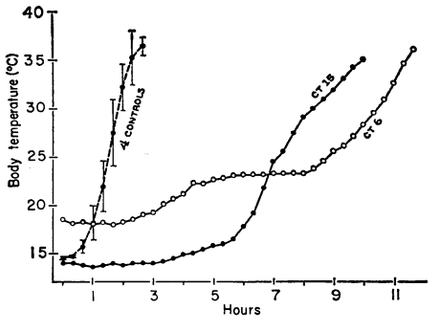

Fig. 1. Time taken by four control and two experimental ground squirrels (CT15 and CT6) to reach a body temperature of at least 35°C. Vertical lines indicate standard error.

from the colony and placed in an individual cage.

One to two weeks after surgery, the squirrels were rendered hypothermic by Giaja's technique of hypercapnic hypoxia (*4*). This entails putting the animal in a closed container at 0°C ambient temperature. As the oxygen in the container is used up and the concentration of carbon dioxide increases, the animal loses its ability to maintain normal body temperature and its temperature falls rapidly. The squirrels were then placed on their backs in large plexiglass cages with wire mesh floors, in a chamber kept at 10°C. Rectal temperatures were taken with a thermistor probe (*5*) inserted 5 cm into the anus and taped to the tail. Recovery rate was determined for three animals at a time, two experimental and one control animal. Their temperatures were recorded every 5 minutes until the last animal had reached a body temperature of at least 35°C, at which point the experiment was terminated.

The experimental animals were anesthetized and killed by perfusion through the heart with saline followed by 10 percent formalin. The brains were removed, frozen, and sectioned coronally at 50 μ. Every alternate section was stained with thionin and mounted on a slide.

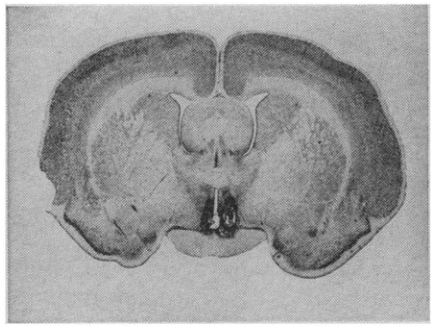

Fig. 2. Photomicrograph of brain of squirrel CT6, showing largest extent of lesions at the level of the anterior commissure and optic chiasm.

When first placed in the recovery chamber, the animals had body temperatures ranging from 14° to 20°C. Figure 1 shows the results for four control animals which started at approximately the same body temperatures. These animals, in an ambient temperature of 10°C, rapidly recovered normal body temperatures. They started in a state of semitorpor, with eyes closed and respiration slow. The first signs of movement were vibrating motions of the front limbs. No righting occurred for a least 15 minutes, nor did their body temperatures rise more than a degree in the first 20 to 40 minutes. After this stage, recovery was rapid. They righted, sat huddled in a ball, and began to shiver. Shivering and respiration rate increased as rewarming progressed and temperatures rose at an average rate of 1°C every 5 minutes, reaching a high rate of 4.5°C in one 5-minute period.

The results for two of the four experimental squirrels which took longest to recover are also shown in Fig. 1. These animals took 10 to 11 hours to reach a body temperature of 35°C in contrast to the four controls which required an average of 2½ hours. Body temperatures of these squirrels fell 0.5°C before beginning to rise. They were observed to shiver, but, since no quantitative measure of shivering was taken, it is not possible to state whether any differences existed between the experimental and control squirrels.

The temperature of another experimental squirrel, which had a rectal temperature of 14.8°C when first placed in the recovery chamber, dropped to 12.5°C in half an hour. To prevent death, a lamp was suspended over it for 15 minutes. This brought its temperature up to 13.5°C, after which the squirrel gradually recovered and reached a temperature of 35°C in 5 hours. At this body temperature all experimental squirrels were active and vocal when handled and appeared normal in all respects.

Squirrel CT6 was particularly interesting because when first placed in the recovery chamber its body temperature was 4°C higher than that of the others shown in Fig. 1. Even so, it took 11 hours to reach 35°C. In general, the higher the starting body temperature, the less time required to reach 35°C. One control squirrel and three squirrels with ineffective lesions started with temperatures of 18° to 19°C and took an average of 106 minutes to recover.

Four of the 16 animals that were operated on were clearly defective in their ability to warm themselves up after they were rendered hypothermic and took far longer, 3½ to 11 hours, to recover. Three of these four had several hypothalamic structures in common that were bilaterally destroyed. The preoptic nuclei were severely damaged—medialis, periventricularis, and pars suprachiasmatic. There was lesser involvement of the anterior hypothalamic nucleus, the suprachiasmatic nucleus, and the periventricular nucleus. A typical lesion, at the level of the anterior commissure, is shown in Fig. 2. In a fourth animal, CT15 in Fig. 1, there was no preoptic damage but almost total destruction of the anterior hypothalamus and suprachiasmatic nuclei. One animal which was indistinguishable from the controls in the time it took to recover had a large lesion in the same area plus slight damage in the medial and periventricular preoptic region. No explanation was found for the lack of the expected deficit in its ability to recover. The other 12 experimental squirrels had either very small preoptic or anterior lesions, or none at all, the electrode placements being too low and in the optic chiasm.

It is clear from this experiment that lesions of the anterior hypothalamic-preoptic region can impair the ability of the thirteen-lined ground squirrel to rewarm itself after induced hypothermia. Therefore, as in homeothermic animals, this area plays an important role in the regulation of body temperature. In addition, it seems reasonable to expect that lesions comparable to those which impair recovery from induced hypothermia should drastically alter the natural process of hibernation.

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References and Notes

1. S. W. Ranson, *Res. Publ. Assoc. Res. Nervous Mental Disease* **20**, 342 (1940).
2. E. Satinoff, *Am. J. Physiol.* **206**, 1389 (1964).
3. C. Frazier, B. Alpers, F. Lewy, *Brain* **59**, 122 (1936).
4. I. Giaja, *Compt. Rend.* **210**, 80 (1940).
5. Yellow Springs Instrument Company.
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