flux. In *Sepia* axons, however, Hodgkin and Keynes (*16*) were not able to influence sodium efflux by passage of hyperpolarizing currents.

The molecular mechanism by which the sodium extrusion system could be influenced by transmembrane voltage is not readily apparent. Conway (*6*) has proposed an oxidation-reduction reaction, as in the cytochrome system, and Kernan (*17*) has found evidence that the cytochrome chain is indeed active during pumping. This suggestion has been criticized on the basis of its energetic inefficiency. It is not difficult to imagine that charged elements in the membrane might be sensitive to electrical potential or that the electrical field in the membrane might influence binding sites for sodium, but our understanding of the sodium pump is inadequate at this time to warrant speculation about its mechanism.

An interesting part of this interpretation that the cell acts to maintain constant its electrochemical gradient for sodium is its voltage dependence. As a result, depolarization alone will activate the pump. An action potential, while produced by a current of sodium ions into the cell, might itself activate the pump. The prolonged action potential of cardiac muscle (250 to 500 msec) should be a good stimulus, and as suggested by Page (*18*) an electrogenic pump might even contribute to the repolarization of the action potential. Little information is available to indicate how rapidly the pump may be stimulated or depressed. Adrian and Slayman (*19*) have found that pump activity appeared to follow temperature changes with a lag of less than 2 seconds, but further discrimination was not possible in their experiments because of the slowness with which the cells were warmed.

HARRY A. FOZZARD
Department of Medicine, University of Chicago, Chicago, Illinois 60637
DAVID M. KIPNIS
Department of Medicine, Washington University Medical School, St. Louis, Missouri

References and Notes

1. A. L. Hodgkin and P. Horowicz, *J. Physiol. London* **148**, 127 (1959).
2. H. H. Ussing, *Physiol. Rev.* **29**, 127 (1949).
3. T. Rosenberg, *Acta Chem. Scand.* **2**, 14 (1948).
4. A. L. Hodgkin, *The Conduction of the Nervous Impulse* (Sherrington Lectures VII) (Thomas, Springfield, Ill., 1964).
5. R. D. Keynes and R. C. Swan, *J. Physiol. London* **147**, 591 (1959); L. J. Mullins and A. S. Frumento, *J. Gen. Physiol.* **46**, 629 (1963); E. J. Harris, *J. Physiol. London* **177**, 355 (1965).
6. E. J. Conway, *Nature* **187**, 394 (1960).
7. ——, R. P. Kernan, J. A. Zadunaisky, *J. Physiol. London* **155**, 263 (1961).
8. H. B. Steinbach, *Am. J. Physiol.* **167**, 284 (1951).
9. P. Horowicz and C. J. Gerber, *J. Gen. Physiol.* **48**, 489 (1965).
10. D. M. Kipnis and C. F. Cori, *J. Biol. Chem.* **234**, 171 (1959).
11. J. E. Parrish and D. M. Kipnis, *J. Clin. Invest.* **43**, 1994 (1964).
12. L. J. Mullins and M. Z. Awad, *J. Gen. Physiol.* **48**, 761 (1965).
13. R. D. Keynes, *J. Physiol. London* **178**, 305 (1965).
14. D. M. Kipnis and J. E. Parrish, *Federation Proc.* **24**, 1051 (1965).
15. H. H. Ussing and K. Zerahn, *Acta Physiol. Scand.* **23**, 110 (1951).
16. A. L. Hodgkin and R. D. Keynes, *Symp. Soc. Exptl. Biol.* **8**, 423 (1954).
17. R. P. Kernan, *J. Physiol. London* **169**, 862 (1963).
18. E. Page and S. R. Storm, *J. Gen. Physiol.* **48**, 957 (1965).
19. R. H. Adrian and C. L. Slayman, *J. Physiol. London* **184**, 970 (1966).
20. Supported by PHS grants HE 09019 and AM 1921.

21 March 1967

Regulation of Body Temperature in the Blue-Tongued Lizard

Abstract. *Lizards* (Tiliqua scincoides) *regulated their internal body temperature by moving back and forth between 15° and 45°C environments to maintain colonic and brain temperatures between 30° and 37°C. A pair of thermodes were implanted across the preoptic region of the brain stem, and a reentrant tube for a thermocouple was implanted in the brain stem. Heating the brain stem to 41°C activated the exit response from the hot environment at a colonic temperature 1° to 2°C lower than normal, whereas cooling the brain stem to 25°C delayed the exit from the hot environment until the colonic temperature was 1° to 2°C higher than normal. The behavioral thermoregulatory responses of this ectotherm appear to be activated by a combination of hypothalamic and other body temperatures.*

Vertebrates regulate internal body temperature by activation of two classes of responses in varying proportions. One class, termed physiological, comprises the secondary functions of organ systems that can modify rates of heat transfer from the core to body surface or from surface to environment, or modify the level of heat generation; shivering, panting, and vasomotor responses are obvious examples. An older class of thermoregulatory responses has been termed behavioral; they are the coordinated activity of the whole animal, selecting or creating a microenvironment in which the optimal internal temperature may be achieved passively. The animal may also modify by behavior one or more of the physical factors effecting heat exchange, such as body shape or orientation, when choice of environment is not possible.

Ectotherms such as lizards (*1*) and fish (*2*) regulate their body temperatures when not constrained from doing so. When a range of thermal environments is available to the animal, it apparently selects one in which a preferred body temperature can be achieved. Although the major thermoregulatory responses in ectotherms are behavioral, certain physiological responses also have been described (*3*). The way in which these responses are activated has not been explored.

We now report that the blue-tongued lizard, *Tiliqua scincoides* (Shaw), also can regulate its body temperature primarily by behavioral responses. Our results suggest that the regulatory responses are activated by a combination of brain and other body temperatures.

A pair of thermodes (1.0 mm in outside diameter, thin-wall, stainless-steel tubes spaced 3 mm apart) were implanted astraddle the brain stem of each of ten lizards. A stainless-steel reentrant tube (0.5-mm outside diameter) was implanted 0.5 mm from the midline and 1.0 mm rostral to the thermodes to accommodate a thermocouple. The temperature of the thermodes was controlled by water from a circulator (*4*) mounted on the lizard's head; water was pumped to and from the circulator through small plastic tubes at 70 ml/min from a constant-temperature bath (45° or 25°C). Three or more weeks after the thermodes were implanted, the lizard was placed in a chamber with a choice of two environments: a dimly lit courtyard at 15°C was surrounded by six aluminum boxes (10 by 20 by 30 cm) heated to 45°C by electrical ribbon heaters which, with thermal insulation, were wrapped around them. The door closed when the lizard entered a heated box, but could be readily opened by the lizard when it was ready to return to the cold courtyard. While the lizard oscillated between the two environments, its colonic, dorsal skin surface, and brain temperatures were continuously recorded by 36-guage, nylon-coated thermocouples (Fig. 1 and Table 1).

When lizard No. 8 entered a hot box, it remained there until its colonic temperature increased to 37.1° ± 1.2°C, when it returned to the cold courtyard until its colonic temperature fell

to 30.0° ± 2.3°C—when it reentered the box. Thus, by activating an appropriate behavioral response, lizards No. 6, 8, and 21 were able to regulate their internal body temperature between 30° and 37°C. Without that response, their body temperatures would have increased to 45°C or decreased to 15°C. Six of the other seven lizards bearing implants also were able to activate this behavioral response in order to regulate their internal body temperature. The seventh made no regulatory response whatsoever when placed in the cold, and in the hot environment its colonic temperature increased to 42°C before it became active; but it never left the hot box by itself. It reacted normally before the implantation and appeared to be normal in all other respects after implantation (that is, it ate well, was alert, and walked normally).

Heating the brain stem to about 41°C, while the lizard was in the hot box, activated the response to exit at a colonic temperature that averaged 2.5°C lower ($p < .01$) than normal for lizard No. 6, 1.4°C lower ($p < .01$) for No. 8, and 1.0°C lower ($p < .01$) for No. 21 in spite of the fact that the surface temperature of the skin also was lower in each instance. On the other hand, cooling the hypothalamus to about 25°C, while the lizard was in the hot box, increased the colonic temperature at exit from the box to 1.4°C above normal ($p < .01$) for lizard No. 6, 2.1°C ($p < .01$) for No. 8, and 0.6°C ($p < .01$) for No. 21 in spite of the fact that the surface temperature of the skin was also higher in each animal.

Thus the brain temperature plays a part in generating the signal to activate the exit response, but the colonic or skin temperature, or both, also must play a part. Heating the brain stem above 40°C did not by itself activate the exit response, but only after the colonic and skin surface temperatures had increased to appropriate values (Fig. 1). Nor did cooling the brain stem indefinitely block the exit response. Only when the colonic and skin temperatures became high enough was the exit response activated, the indication being that these temperatures also must be involved in generation of the activating signal.

The effects of heating and cooling the brain stem upon the colonic temperature at exit from the cold to the hot environment were not so clearly demonstrated as were these effects upon the colonic temperature at exit from the hot to the cold environment. Heating the brain stem did decrease the colonic temperature at exit from the cold environment in lizard No. 6, but not in No. 8 or No. 21. Cooling of the brain stem increased above normal the colonic temperature at exit from the cold environment in lizards No. 8 and 21, but not in No. 6.

The design of the experiment was not suitable for precise determination of the limits of the preferred temperature range—especially the lower limit.

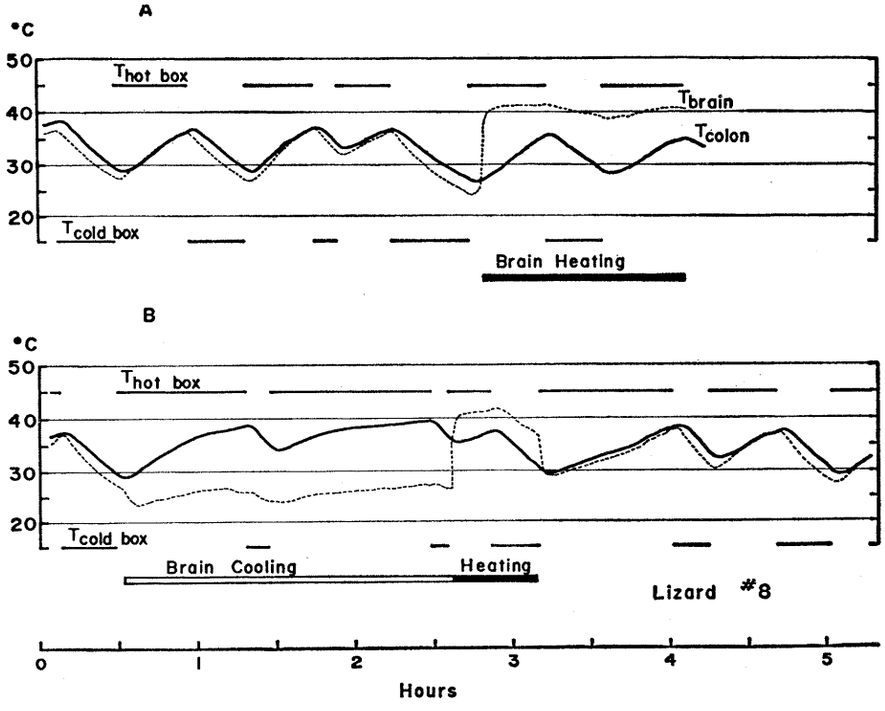

Fig. 1. Colonic and brain-stem temperatures of lizard No. 8 (body weight, 625 g) while it oscillated between a hot and a cold environment. The brain temperature was normal except where heating or cooling of the brain with thermodes is indicated.

Table 1. Mean (± S.D.) body temperatures of three lizards at time of movement from hot (45°C) to cold (15°C) and from cold to hot environments. Numbers of observations appear in parentheses.

Lizard (No.)	Temperature source	Thermal state of brain stem	Temperature (°C) at time of passage from: Heat to cold		Cold to heat	
6	Colon	Normal	36.7 ± 1.3	(21)	31.0 ± 2.5	(22)
6	Colon	Heated	34.2 ± 2.0*	(16)	24.1 ± 1.9*	(5)
6	Colon	Cooled	38.1 ± 1.2†	(5)	31.3 ± 3.7	(15)
6	Skin	Normal	36.0 ± 1.0	(21)	28.0 ± 2.4	(22)
6	Skin	Heated	33.3 ± 1.8*	(16)	22.3 ± 1.9*	(5)
6	Skin	Cooled	37.3 ± 1.1*	(5)	28.3 ± 2.9	(15)
6	Hypothalamus	Normal	35.5 ± 1.5	(20)	30.3 ± 2.3	(22)
6	Hypothalamus	Heated	40.3 ± 0.5	(16)	38.2 ± 0.2	(5)
6	Hypothalamus	Cooled	24.6 ± 3.7	(5)	25.0 ± 2.0	(15)
8	Colon	Normal	37.1 ± 1.2	(22)	30.0 ± 2.3	(23)
8	Colon	Heated	35.7 ± 0.7*	(8)	30.0 ± 0.8	(3)
8	Colon	Cooled	39.2 ± 0.7*	(5)	32.6 ± 3.3	(6)
8	Skin	Normal	37.0 ± 1.1	(22)	26.2 ± 1.9	(23)
8	Skin	Heated	36.0 ± 1.0†	(8)	25.5 ± 0.9	(3)
8	Skin	Cooled	38.5 ± 0.8*	(5)	28.3 ± 2.8†	(6)
8	Hypothalamus	Normal	36.2 ± 1.3	(22)	28.2 ± 2.0	(23)
8	Hypothalamus	Heated	40.7 ± 1.0	(8)	38.2 ± 1.4	(3)
8	Hypothalamus	Cooled	26.0 ± 1.9	(5)	23.0 ± 3.2	(6)
21	Colon	Normal	37.4 ± 0.6	(20)	29.8 ± 3.2	(20)
21	Colon	Heated	36.4 ± 0.7*	(15)	32.5 ± 1.2	(10)
21	Colon	Cooled	38.0 ± 1.3*	(6)	31.5 ± 2.3	(9)
21	Skin	Normal	36.8 ± 1.1	(20)	25.2 ± 1.8	(20)
21	Skin	Heated	35.7 ± 1.4*	(15)	27.0 ± 1.5	(10)
21	Skin	Cooled	37.2 ± 1.3	(6)	27.3 ± 2.6*	(9)
21	Hypothalamus	Normal	36.5 ± 0.8	(20)	28.8 ± 3.1	(20)
21	Hypothalamus	Heated	41.4 ± 0.5	(15)	40.5 ± 0.5	(10)
21	Hypothalamus	Cooled	25.5 ± 3.3	(6)	23.5 ± 2.3	(9)

* $p < .01$. † $p < .025$.

When the animal learned that the only choice was between an environment that was too hot and one that was too cold, its decision to leave the cold environment may have been influenced by remembrance that the other environment would shortly become intolerably hot. Remaining in the cold courtyard would in no way be harmful, whereas an extended period in the hot environment could be lethal. Drowsiness and sleep sometimes appeared to depress the response to enter the hot box, especially when the brain stem was heated. If the body temperature fell below about 25°C or if it was below this level at the start, our lizards seldom entered the hot box unassisted. Cold-induced lethargy did not immobilize them, however, as they could easily be prodded to move at a body temperature of 15°C.

In all lizards except No. 8 (still alive 4 months after implantation), the placement of the thermodes was determined. The thermodes were straddling the optic chiasm and the preoptic region above it in lizards No. 6 and 21. In the others, the thermodes were 1.0 to 2.0 mm caudal to the optic chiasm; the effect of heating or cooling the brain stem was less than in lizards No. 6, 8, and 21. Thus there is a suggestion that the preoptic region is the thermally responsive region of the brain stem. In endotherms this same region is known to activate physiologic thermoregulatory responses when its temperature is displaced.

Bartholomew *et al.* (5), placing two lizards (*T. scincoides*) in a 40°C environment with a starting body temperature of 20°C, obtained heating curves; and in a 20°C environment, with a starting body temperature of 40°C, they obtained cooling curves. The rate of heating, at a body temperature of 30°C, was greater than the rate of cooling at the same temperature; the difference was attributed to the greater heat production during heating, so that the calculated conductance of the tissue and air, between the core temperature and environment, was the same for both heating and cooling. Their results probably mean that any circulatory adjustments that may be associated with body temperature were not shown to have thermoregulatory significance. We arrived at the same conclusion, since heating or cooling of the preoptic region of lizard No. 21 had no discernible effect upon either the heating or the cooling curves for this animal. Nor did we see any inflections in the normal heating or cooling curves (brain stem not heated or cooled) to suggest a change in blood flow from core to skin.

However, the heat capacity of animal tissue is great, so a method based upon heating or cooling curves is not sufficiently sensitive for detecting small thermoregulatory responses. We noticed that the vessels along the margin of the ventral scales are engorged with blood when the animal is hot (35°C), but we found that heating or cooling of the brain stem had no effect on the amount of blood in these vessels or on their rate of filling after they were emptied by gentle pressure.

Hyperventilation and gaping have been observed in heated lizards (*6*), and these we observed in *T. scincoides*; however, the associated loss of evaporative water had no discernible effect on the heating curves, nor did heating or cooling of the hypothalamus indirectly affect the heating curves by affecting the evaporative heat loss from the mouth. The only effect observed when the brain stem was heated while a heating curve was recorded was that the lizard struggled more vigorously and at a lower colonic temperature. Likewise, cooling of the brain while body temperatures were rising delayed the struggling until a higher colonic temperature was achieved.

H. T. HAMMEL
FRED T. CALDWELL, JR.
ROBERT M. ABRAMS
John B. Pierce Foundation Laboratory, New Haven, Connecticut, and *Upstate Medical Center, State University of New York, Syracuse*

References and Notes

1. R. B. Cowles and C. M. Bogert, *Bull. Amer. Museum Nat. Hist.* **83**, 261 (1944); C. M. Bogert, *Evolution* **3**, 195 (1949); K. S. Norris, *Ecology* **34**, 265 (1953); O. P. Pearson, *Copeia* **1954**, 111 (1954); H. Saint-Girons and M. C. Saint-Girons, *Vie Milieu* **7**, 133 (1956); J. E. Heath, *Univ. Calif. Publ. Zool.* **64**, 97 (1965).
2. F. E. J. Fry, *Publ. Ontario Fisheries Res. Lab.* **55**, 5 (1947); T. K. Pitt, E. T. Garside, R. L. Hepburn, *Can. J. Zool.* **34**, 555 (1956); P. N. Rozin and J. Mayer, *Science* **134**, 942 (1961).
3. W. R. Dawson and J. R. Templeton, *Physiol. Zool.* **36**, 219 (1963); V. H. Hutchison, H. G. Dowling, A. Vinegar, *Science* **151**, 694 (1966).
4. Similar in design to one constructed for use with dogs and ground squirrels; H. T. Hammel, *Univ. Missouri Agr. Exp. Sta. Spec. Rep. 73* (1966).
5. G. A. Bartholomew, V. A. Tucker, A. K. Lee, *Copeia* **1965**(2), 169 (1965).
6. W. R. Dawson and G. A. Bartholomew, *Physiol. Zool.* **31**, 100 (1958).
7. Supported by NSF grant GB-4259. The study was part of the program for Operation Billabong and was conducted aboard R.V. *Alpha Helix* (Scripps Institution of Oceanography) at the Great Barrier Reef during August and September 1966.

1 February; revised 31 March 1967

Ischemic Necrosis: Prevention by Stress

Abstract. *Complete interruption of the circulation, by means of a special clip, in a skin flap during 9 hours results in necrosis of the ischemic area. Prior treatment with various severe stressors (spinal-cord transection, prolonged restraint, quadriplegia due to transection of motor nerves, forced exercise, or cold baths), as well as systemic injection of epinephrine, norepinephrine, or chlorpromazine, offers virtually complete protection against this form of topical tissue injury.*

One hundred twenty female Sprague-Dawley rats with an average body weight of 100 g (range, 90 to 110 g) were separated into 12 equal groups and treated as indicated in Table 1.

Table 1. Prevention of necrosis of the skin by stress. In addition to the treatments listed, a skin flap was isolated from the circulation by a compressing clip as indicated in the text. All subcutaneous injections were administered on the abdomen. Times (minutes, hours, days) preceded by minus signs are the lengths of time before application of the treatment; plus signs, after treatment.

Treatment	Cutaneous necrosis (scale, + to +++)
None	+++
Spinal-cord transection	
Thermocautery between C7 and D1 at 0 hour	0
Restraint	
24 hours on a board, beginning at −15 hours	±
Quadriplegia	
Transection of motor nerves of all four extremities at −1 hour	+
Starvation	
Only water allowed from −48 hours to +9 hours	+
Forced exercise	
In drum, 30 cm in diameter, 12 rev/min, from −30 minutes to +6 hours and from +8 hours 30 minutes to +9 hours	+
Cold	
Kept at 9°C from −30 minutes to +9 hours	Trace
Epinephrine	
Subcutaneous injection of 0.8 mg at −30 minutes	Trace
Norepinephrine	
Subcutaneous injection of 1.5 mg at −30 minutes	0
Cortisol acetate	
Daily subcutaneous injection of 3 mg from −5th day to +1st day	+++
Cortisol sodium succinate	
Intravenous injection of 20 mg at −30 minutes	+++
Chlorpromazine	
Subcutaneous injection of 1.5 mg at −30 minutes	Trace