

As shown in Figure 2d, electrical stimulation with 2–4 times threshold intensity produced the second group of action potentials (II), which exhibited long and fluctuating latency. The minimum latency of the second action potentials, corrected for conduction time, was a mean of 5.1 msec (26 units) with a mean fluctuation range of 4.9 msec. Similar values have been obtained in the frog water-sensitive fibres by NOMURA and KATSUHATA¹³. Since the 5 msec latency of the second impulses in response to electrical stimulation is much shorter than the 86 msec latency to a 1.0 M NaCl stimulus, it seems unlikely that the second impulses were induced by gustatory stimulation of the taste receptor sites with electrophoretically carried Ringer ions at the tip of Ringer-filled suction electrode. The second fluctuating impulses completely disappeared with repetitive electrical stimulation at 10 Hz. Even when the repetition rate was 1 Hz, the responses occurred intermittently, as shown in Figure 2d. Because of instability at low frequencies of stimulation, it is not considered that the second group of action potentials was generated by direct electrical stimulation of gustatory nerve terminals. When the taste bud located at the summit of the fungiform papilla was mechanically destroyed, no second impulse after the first impulse was observed. Therefore, it is concluded that the second impulses with irregular latencies originated synaptically from the result of direct depolarization of taste cell membranes by electrical current. With neuromuscular junctions, it is well known that repetitive electrical stimulation of the pre-synaptic axon with low frequency produces endplate potentials with fluctuating latency and intermittent oc-

currence¹⁴. These properties are very similar to those of the second group of gustatory impulses under the present investigation.

Although the whole time course of transduction steps of a gustatory stimulus into nerve signals is as yet unknown, the present experiment indicates that the total time interval between the onset of taste cell depolarization and the initiation of gustatory neural impulse is about 5 msec. This indicates that about 94% of an 86 msec latency produced by 1.0 M NaCl is the time between the onset of gustatory stimulation and the generation of the taste receptor potential, i.e., the latency of receptor potential. Thus, most of the portion of the latency of NaCl-induced taste nerve responses may be due to the time required for effective adsorption of taste stimuli on the taste receptor membrane², because the latency is greatly dependent on taste stimulus concentrations as shown in Figure 1d.

SATO⁵ has already shown that the time between the presentation of 0.5 M NaCl and the onset of intracellular receptor potential of frog taste cells was 100–300 msec. These values are similar to those seen in Figure 1c and d. Therefore, it is concluded that the most important factor determining the latency of gustatory neural impulses is the latency of taste cell depolarization following initiation of taste stimulation.

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¹⁴ B. KATZ and R. MILEDI, *Proc. R. Soc. B* 167, 23 (1967).

Spontaneous Thermoregulatory Oscillations in Cutaneous Efferent Sympathetic Activity

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Summary. Patterns of changes in cutaneous efferent sympathetic activity which have previously been shown to occur during experimental manipulation of the temperature of various thermosensitive body sites, have now been shown to accompany spontaneous thermoregulatory activity. That is, under thermoneural conditions, concurrent spontaneous oscillations in skin blood flow and efferent sympathetic activity were observed.

Patterns of efferent sympathetic activity appropriate to regional circulatory responses elicited by changes in temperature of the hypothalamus, spinal cord or skin have recently been clearly demonstrated^{2–4}. Such work has involved specific experimental manipulation of the temperature of an area of skin, the hypothalamus or spinal cord, and although normal thermoregulatory effector mechanisms accompany such changes (see reviews by HALES⁵ and SIMON⁶), the accompaniment of spontaneous thermoregulatory activity by appropriate efferent sympathetic activity has not been reported; this is, of course, essential if proposed mechanisms are to be accepted as entirely valid, and the present study has examined this.

Methods. Observations have been made on 3 albino rabbits of either sex, weighing 2–3 kg; they were artificially ventilated while anaesthetized with sodium pentobarbital (initial dose of 30 mg kg⁻¹, followed by an infusion of 4 mg kg⁻¹ h⁻¹) and immobilized with succinylcholine (20 mg initially, followed by an infusion of 100 µg kg⁻¹ h⁻¹). Polyethylene thermodes were surgically placed in the vertebral canal, extending from the lower lumbar to the mid-cervical level.

Continuous monitoring was made of the skin temperature of both ears, rectum, spinal canal and ambient air, and of the electrical activity of a postganglionic nerve twig accompanying one of the retroauricular arteries (cutaneous 'ear' sympathetic). The nerve potentials were recorded directly and also after integration over 4 sec intervals. Full details of these techniques have been given previously^{2–4}.

At the beginning of an experimental period, the rabbit was placed on its side on a water perfused plate, and water and ambient air temperatures were adjusted until ear skin temperatures lay approximately mid-way between

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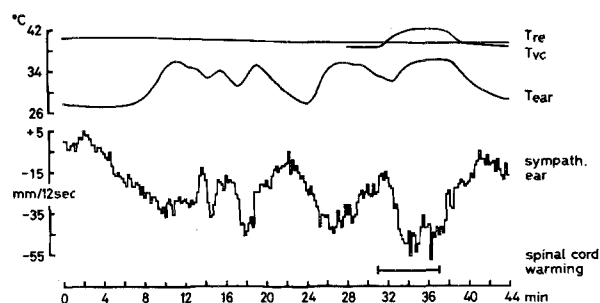
⁴ W. RIEDEL, M. IRIKI and E. SIMON, *Pflügers Arch.* 332, 239 (1972).

⁵ J. R. S. HALES, in *MTP International Review of Science* (Ed. D. ROBERTSHAW; Butterworths, London 1974), Series 1, vol. 7, p. 107.

⁶ E. SIMON, *Rev. Physiol. Biochem. Pharmac.* 71, 1 (1975).

rectal and air temperature; water and air temperatures were then maintained constant. For comparative purposes, the effects of warming the spinal cord to 41–42°C were also observed.

Results and discussion. Efferent sympathetic activity and the various temperatures were monitored for up to 8 h. The results presented in the Figure show that even with rectal, plate and air temperatures essentially constant, spontaneous oscillations in ear skin temperature occur and are accompanied by inverse changes in the level



Integrated cutaneous efferent sympathetic activity (sympath. ear), and temperatures of the ear (T_{ear}), vertebral canal (T_{vc}) and rectum (T_{re}) of a rabbit lying on a pad at 37.8–38.1°C with ambient dry bulb temperature 23.8–25.1°C.

of electrical activity of the nerve. Ambient air temperature was usually about 25°C, plate water 40°C, and rectal temperature about 39.5°C. Ear skin temperature varied by up to 8°C, while nerve activity changed by as much as 90% of the difference in level of activity seen in the constricted and fully dilated states; the duration of oscillations was 2–70 min. Warming the spinal cord could result in complete abolition of spike activity in excess of the baseline noise level, and was associated with a 0.5–8°C increase in ear skin temperature over a period of approximately 6 min.

The observed changes in ear skin temperature are indicative of oscillations in blood flow. The spontaneous nature of these events is characteristic of the activity of thermoregulatory mechanisms when an animal is in the thermoneutral zone. At this time, thermoregulatory effector mechanisms exhibit minimal activity, and the fine adjustment of body temperature is brought about by small adjustments in cutaneous blood flow (e.g., see symposium edited by MONTEITH and MOUNT⁷). The spontaneous oscillations in sympathetic activity controlling the cutaneous blood flow indicate that the previously reported changes which were experimentally evoked^{2–4} represent normal physiological phenomena.

⁷ J. L. MONTEITH and L. E. MOUNT, Symposium: *Heat Loss in Animals and Man* (Butterworths, London 1974).

Changes in the Free Amino Acid Pattern of Haemolymph of the Common Indian Scorpion *Palamnaeus bengalensis* During Moulting

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Summary. Haemolymph of *Palamnaeus bengalensis* contains normally 12 and during moulting up to 15 free amino acids, being in maximal number at the end of ecdysis. Aminoacidaemia is most pronounced during the pharate stage. Tyrosine appeared for tanning of the cuticle. Taurine and methionine were not present.

SHRIVASTAVA^{2,3} described structure and histochemical composition of the various layers of cuticle of *P. bengalensis* in the different stages of their development during moulting. In the present study, corresponding changes in the free amino acid pattern of *P. bengalensis* during different stages of moulting have been studied. Though data is available on free amino-acids (FAA) in insects, much less is available in arachnids^{4–8} and none on the changes of FAA during moulting of scorpions.

Materials and methods. Known quantities of haemolymph samples of *P. bengalensis* in different stages of moulting were collected and extracted in ethanol for their amino acids which were detected by thin layer chromatography and the two dimensional paper partition chromatography. Standard solutions of pure amino-acids prepared in 10% isopropanol were used for the identification and quantitative assessment.

Observations and discussion. Hemolymph of *P. bengalensis* was found to contain normally 12 and during a moulting period up to 15 free amino acids (Table). The analyses also revealed remarkable variations in their pattern during different stages of moulting.

At premoulting stage, these were present in larger quantity and in greater number than when moulting was in progress, apparently due to the dissolution of some of the old cuticle. Decrease of amino-acids while moulting was in progress was likewise due to their incorporation in

the new cuticle. Then increase to a maximum number at the end of ecdysis shows that some are added from the body.

Seven amino-acids, namely alanine, arginine, aspartic acid, glutamic acid, its amide glutamine, glycine, and tyrosine, were present during all the stages of moulting and the intermolt, but they also showed decline after the ecdysis. Decrease in aspartic acid, leucine, lysine, phenylalanine, threonine, tryptophan, and valine appeared to be related to increase in body size of the scorpion, taking place during this period, and decrease of arginine, glycine, phenylalanine, proline, tryptophan and tyrosine was related specially to moulting.

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