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THE CENTRAL CONTROL OF SHIVERING AND NON-SHIVERING THERMOGENESIS IN THE RAT

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SUMMARY

1. To test whether the preoptic area controls only non-shivering and the spinal cord only shivering thermogenesis, ten rats were chronically implanted with a preoptic and a spinal cord thermode each. The following were then studied: (*a*) the effect of propranolol (8 mg/kg.hr) on the metabolic response to cooling the preoptic area, and the spinal cord, (*b*) the effect of exogenous noradrenaline (0·5 mg/kg) on the metabolic response to cooling the preoptic area, and the spinal cord, and (*c*) the effect of warming the preoptic area on the metabolic response to cooling the spinal cord, and vice versa.

2. Administration of propranolol inhibited the metabolic response to cooling each of the thermosensitive areas, but the response to cooling the preoptic area was more strongly inhibited than that to cooling the spinal cord.

3. Administration of exogenous noradrenaline did not prevent the metabolic response to cooling either the preoptic area or the spinal cord.

4. Warming the spinal cord completely inhibited the metabolic response to cooling the preoptic area, and warming the preoptic area fully inhibited the metabolic response to cooling the spinal cord.

5. It is concluded that exogenous noradrenaline underestimates the capacity for non-shivering thermogenesis, and that both thermosensitive areas can control both forms of thermogenesis, but that the preoptic area threshold of non-shivering thermogenesis is probably lower than that of shivering, while the spinal cord threshold of shivering is probably lower than that of non-shivering thermogenesis.

INTRODUCTION

It is well known that many mammals, but particularly small ones, possess two forms of thermoregulatory heat production: shivering, which is a skeletal muscle tremor, and non-shivering thermogenesis which, by definition, is any thermoregulatory thermogenic process that does not involve skeletal muscle contraction. These two forms of heat production are under the control of the hypothalamic temperature regulation centre; shivering is mediated by the somatic motor efferents and there is little doubt that non-shivering thermogenesis, no matter what the effector organs are, is mediated by the sympathetic nervous system because it can be blocked by ganglionic (Hsieh, Carlson & Gray, 1957) and adrenergic beta-receptor

(Brück & Wünnenberg, 1965) blocking agents, and stimulated by noradrenaline (Hsieh & Carlson, 1957; Hsieh *et al.* 1957).

The control of total thermoregulatory heat production has evoked great interest in the last two decades but the selective control of each form of thermogenesis has received little attention. The available evidence in the new-born and in the cold adapted guinea-pig (Brück & Wünnenberg, 1970), as well as in the cold exposed rat (Fuller, Horwitz & Horowitz, 1975; Fuller, Horowitz & Horwitz, 1977), suggests that shivering is controlled by cutaneous and spinal cord thermal sensors, while non-shivering thermogenesis is controlled by cutaneous and hypothalamic thermal sensors. According to this dual control system, cold exposure elicits first non-shivering thermogenesis because the heat produced in the cervical brown adipose tissue warms the spinal cord and, therefore, counteracts the cutaneous drive for shivering (Brück & Wünnenberg, 1970). Shivering, thus, will only be elicited when the capacity for non-shivering thermogenesis has been more or less used-up and the temperature of the spinal cord decreases below the threshold of shivering.

This dual control system, however, does not explain a number of observations in rats adapted to ambient temperatures close to thermoneutrality. Local cooling of the preoptic area, for example, increased resting oxygen consumption by about 150 % (Banet & Hensel, 1976*a*), though the capacity for non-shivering thermogenesis of these animals, as estimated by the administration of noradrenaline (Janský, 1973), was only 33 % of their resting metabolic rate. This metabolic response, therefore, suggests that shivering had been induced by cooling the preoptic area and, thus, supports the several reports of shivering upon cooling of this central thermoreceptive area in the rat (Reichlin, 1964; Satinoff, 1964; Murgatroyd & Hardy, 1970; Stoner, 1972; Banet & Hensel, 1976*a*). On the other hand, some experiments also suggest that the thermosensors in the spinal cord can control non-shivering thermogenesis. Warming the spinal cord, for example, suppressed the increase in oxygen consumption induced by exposure to mild cold stress (Banet & Hensel, 1976*c*). This must have been due to the suppression of non-shivering thermogenesis since in the rat (Banet & Hensel, 1977) and in the guinea-pig (Brück & Wünnenberg, 1970) cold exposure first induces this form of heat production. Furthermore, prolonged and repetitive cooling of the spinal cord of the rat led to an increase in the capacity for non-shivering thermogenesis (Banet & Hensel, 1976*b*).

In view of the above observations, a closer look at the central control of shivering and non-shivering thermogenesis in the rat was clearly necessary. In this study, we have examined, by three independent tests, the working hypothesis that the hypothalamic sensors can only control non-shivering thermogenesis, while those in the spinal cord can only control shivering. If this hypothesis is correct, and providing the experimental manipulation of local hypothalamic or spinal cord temperatures does not induce changes in core or skin temperature that could effect the results, then the following three predictions can be made.

(*a*) Administration of propranolol should prevent the increase in oxygen consumption elicited by cooling the preoptic area, but should have no effect on that induced by cooling the spinal cord, since propranolol is an adrenergic beta-receptor blocking agent which blocks non-shivering thermogenesis but is generally thought not to interfere with shivering (Brück & Wünnenberg, 1965; Schönbaum, Johnson, Sellers

& Gill, 1966; Heim & Hull, 1966; Alexander & Williams, 1968; Blatteis, 1976; Horwitz & Hanes, 1976; Banet & Hensel, 1977) or the cardiovascular system (Brück & Wünnenberg, 1965; Heim & Hull, 1966; Banet & Hensel, 1977).

(*b*) Administration of noradrenaline should also more or less prevent the increase in oxygen consumption elicited by cooling the preoptic area because this exogenous noradrenaline would saturate the adrenergic receptors that control non-shivering thermogenesis, but should have little effect on the increase in oxygen consumption elicited by cooling the spinal cord.

(*c*) Warming the hypothalamus should have little or no effect on the increase in oxygen consumption elicited by cooling the spinal cord, and warming the spinal cord should also have little or no effect on the increase in oxygen consumption elicited by cooling the hypothalamus.

The present results show that these predictions are not borne out and that the working hypothesis is therefore untenable.

METHODS

A group of white male rats were housed at 23 °C, with artificial illumination and a day–night cycle of 12 hr. Food and water were continuously available. The animals were anaesthetized with 45 mg sodium pentobarbitone/kg and the following things were done to each animal: (*a*) a water-perfused thermode (Banet & Seguín, 1967) was implanted stereotaxically into the preoptic-anterior hypothalamic area using the De Groot's atlas (1959) and co-ordinates of 2·2 mm anterior to bregma, 8–8·5 mm below the surface of the cortex and within 1 mm of the mid line; (*b*) a water-perfused thermode made of a length of PE 10 polyethylene tubing bent into a tight U shape (Banet & Hensel, 1976*b*) was implanted into the cervicothoracic spinal canal; and (*c*) a length of PE 10 polyethylene tubing was implanted into the abdominal cavity and the free end brought subcutaneously to the head; the thermodes and the free end of the abdominal tube, used for I.P. administration of propranolol and saline, were then fixed to the calvarium with three stainless-steel screws and dental acrylic.

In the days following the operation, the animals that showed no signs of motor impairment were extensively trained to the experimental procedure. It was of importance to the subsequent experiments that the animals could be trained to remain very quiet when fixed by the tail in a holder and when the thermodes were perfused by water. Some of the animals could not be trained to the required level, while others developed late motor impairment probably due to a displacement of the spinal thermode. These were discarded. Of the animals that remained, only six could be used in all the experiments, while another four, because of damage to one of their thermodes, could only be used in some of them.

At the beginning of all the experiments, each animal was fixed by the tail to a holder that restricted its capacity to turn around. One thermocouple was then inserted about 50 mm beyond the anus, another was applied to the tail skin about 10 mm from its base, both being fixed to this structure, and a third one was fixed to a shaved area in the midline of the dorsal surface of the abdomen. As required, the thermodes were connected to temperature controlled baths and the abdominal tube to an infusion pump. The animal was then placed in a 5 l., temperature controlled chamber at 24–25 °C the air of which was renewed at the rate of 1 l./min, the exhaust air being passed through a paramagnetic oxygen analyser. The following experiments were then done, generally but not always, in the order in which they are presented.

(*a*) *Effect of propranolol on the metabolic response to cooling the preoptic area, and the spinal cord.* Shortly after the animals were placed in the chamber, 1 ml. saline/kg was injected I.P. and, thereafter, saline was continuously infused I.P. at the rate of 1 ml./kg.hr for the duration of the experiment. After 1 hr of infusion, which was the time allowed for the animal to reach thermal equilibrium, a control period of 15–20 min began. At the end of this period, the hypothalamic thermode was perfused with water at a certain temperature during 20 min. The perfusion was then discontinued and, when the parameters measured had more or less stabilized, the thermode was again perfused with water at another temperature. This procedure was

repeated until the thermode had been perfused in random order with water at 38, 30, 20 and 10 °C. During the whole experiment all parameters were measured at 5 min intervals.

At least 24 hr afterwards, the same protocol was repeated, normally at the same time of the day, the only difference being that the animals received propranolol instead of saline, 8 mg/kg at the beginning of, and 8 mg/kg.hr during the experiment. The solution of propranolol in saline, 8 mg/ml., was prepared each day.

Some days afterwards, the whole protocol was repeated but this time the spinal cord thermode was perfused instead of the hypothalamic one.

This series of experiments was completed in six animals, while in another three animals it was completed giving 20 mg propranolol/kg, or an equivalent volume of saline, and continuously infusing the same amount per hour.

(*b*) *Effect of exogenous noradrenaline on the metabolic response to cooling the preoptic area, and the spinal cord.* On this series of experiments, the animals were placed in the temperature controlled chamber for at least 1 hr, which was followed by a 15 min control period. The animals were then removed from the chamber, but not from the holder, to be injected I.M. with 0·5 mg noradrenaline/kg. Immediately after the injection, the animals were replaced in the chamber and all parameters were measured, as before the injection, at 5 min intervals.

At least 3 days later, the protocol was repeated, the only difference being that 15 min after the injection of noradrenaline either the preoptic area or the spinal cord thermode was perfused with water at 10 °C during 10 min. The protocol was again repeated a few days later, so that, both thermoreceptive areas were once cooled in each animal.

This series of experiments was completed in seven animals.

(*c*) *Effect of warming the preoptic area on the metabolic response to cooling the spinal cord, and vice versa.* These experiments, like the previous ones, began with 1 hr for equilibration followed by a 15 min control period. Either the preoptic or the spinal cord thermode was then perfused with water at 30 °C. When oxygen uptake had reached steady state, the other thermode was perfused with warm water (preoptic 45 °C; spinal cord 47 °C) for 15–25 min. The central warm stimulation was then discontinued, but the animals were still stimulated by the cold thermode for 15–20 min, and then the central cold stimulation was also discontinued. All parameters, measured throughout the experiment at 5 min intervals, were followed for a further 10–15 min. At least 24 hr later, this protocol was repeated cooling this time the thermoreceptive area that had been warmed before, and vice versa.

This series of experiments was completed in eight animals.

Throughout the paper, the standard error is used as a measure of dispersion and the average changes in the various parameters were always compared by *t* tests for paired data.

RESULTS

At the beginning of the experiments, the animals weighed 306 ± 14 g. Thereafter they grew steadily but slower than non-operated animals. Throughout the results, oxygen consumption will be expressed as a function of body weight using the standard interspecies exponent of 0·73 (Brody, 1945).

(*a*) *Effect of propranolol on the metabolic response to cooling the preoptic area, and the spinal cord.* To illustrate the experimental procedure, Fig. 1 presents the results of one experiment in which the preoptic area was cooled and saline continuously infused. Note that oxygen consumption first increased but then, as the central cold drive was counteracted by the increased core and furred skin temperatures, it dropped sharply. Cooling the spinal cord (not shown in the Figure), however, induced little change in rectal and furred skin temperatures, while oxygen consumption increased steadily reaching a plateau after 15 min of cooling.

In these series of experiments, we wanted to measure the maximum metabolic response to cooling the preoptic area, and the spinal cord. At each preoptic thermode temperature, therefore, oxygen consumption and the other parameters were taken as the average of the values 10 and 15 min after the perfusion of the thermode began. At each spinal cord thermode

temperature, the values of 15 and 20 min after the beginning of the perfusion were used, however, because only then was the maximum metabolic effect reached. The average of the values in the last 10 min of the control period differed neither from those measured during the 10 min preceding each change in thermode temperature nor from those measured at a thermode temperature of 38 °C. These values at 38 °C thermode temperature were, therefore, used as control.

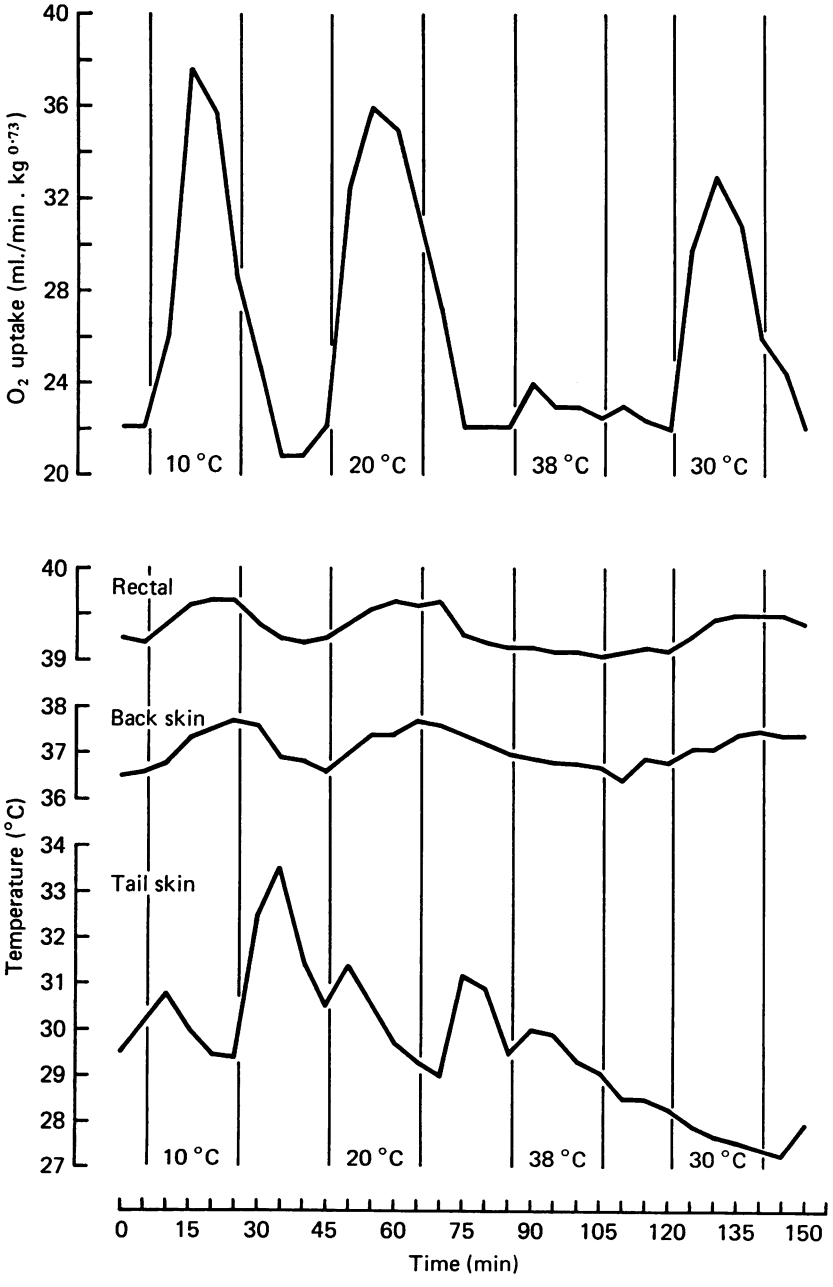

Fig. 1. Effect of cooling the preoptic area on oxygen consumption and various body temperatures in one rat during exposure to 24–25 °C. The vertical lines show the beginning and the end of the perfusion of the thermode with water at the temperatures shown.

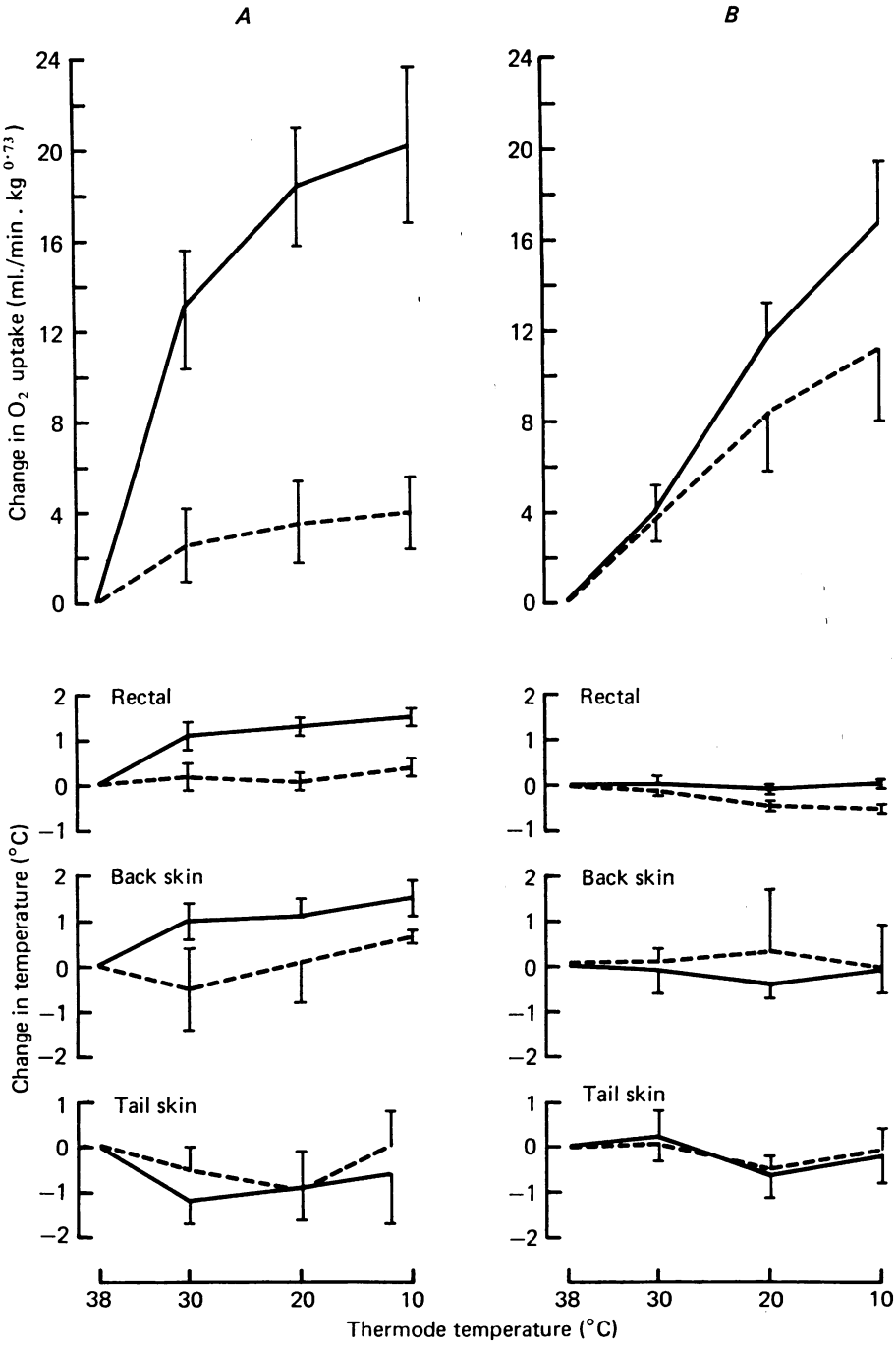

Fig. 2. Average changes in oxygen consumption and various body temperatures induced by cooling the preoptic area (left side of the Figure, *A*) and the spinal cord (right side of the Figure, *B*) in six rats exposed to 24–25 °C during infusion of saline (continuous line) and propranolol (interrupted line). The vertical lines are standard errors.

Resting oxygen consumption and rectal temperature (at a thermode temperature of 38 °C) were $17{\cdot}1 \pm 1{\cdot}6$ ml./min.kg$^{0{\cdot}73}$ and $37{\cdot}9 \pm 0{\cdot}2$ °C, respectively. Administration of propranolol induced a non-significant fall in oxygen consumption of 0·6 ml./min.kg$^{0{\cdot}73}$, but the fall in rectal temperature of 0·7 °C was probably significant ($P < 0{\cdot}05$).

The changes induced in all parameters by cooling each of both central thermosensitive areas, with and without administration of propranolol, are shown in Fig. 2. Cooling the preoptic area (Fig. 2*A*) induced significant increases in oxygen uptake, rectal and back skin temperatures; tail skin temperature, however, fell slightly but the fall was not significant. All these effects were greatly curtailed by the administration of propranolol. After administration of this drug, at a thermode temperature of 10 °C, back skin temperature increased significantly ($P < 0{\cdot}01$) and oxygen consumption probably too ($P < 0{\cdot}05$). None of the other changes in any of the parameters were statistically significant. Thus, propranolol did not completely inhibit the increase in metabolic rate induced by cooling the preoptic area and this metabolic increase was mediated by the thermosensors in this area because the slight fall in unfurred skin temperature, which could have driven the response, was certainly counteracted by the increases in core and furred skin temperatures.

Cooling the spinal cord (Fig. 2*B*) significantly increased oxygen consumption but had no significant effect on the other parameters. After administration of propranolol, however, rectal temperature fell significantly at thermode temperatures of 20 and 10 °C and, at these thermode temperatures, oxygen consumption did not increase as much as before. It is significant that propranolol decreased the metabolic response to cooling the spinal cord and that this effect could not have been mediated by any other thermosensitive area because skin temperature was not affected by cooling the spinal cord and core temperature fell, which would induce an increase rather than a decrease in metabolic rate.

During these experiments, shivering could be clearly observed at the lower spinal cord thermode temperatures both before and during propranolol injection and infusion; when the preoptic area was cooled, however, only a fine tremor could occasionally be observed. This muscle tremor during preoptic cooling was further studied by palpation of the muscles of the neck in which shivering is most intense in the rat (Hart, 1971). It is known that a rat may tremble during even the slightest manual restraint even at ambient temperatures above thermoneutrality, but at the end of this series of experiments the animals were so used to being handled that no tremor could be felt even at normal room temperature which is below thermoneutrality. In five of the rats, perfusion of the preoptic thermode with water at 10 °C elicited a clear tremor with the typical waxing and wanning of shivering, tremor that disappeared when the perfusion of the thermode was discontinued.

In three animals, not used for the above experiments, the protocol was repeated but giving an initial dose of propranolol of 20 mg/kg and the same amount being continuously infused every hour. In these animals, the results were essentially the same as above. At 10 °C preoptic thermode temperature, oxygen consumption increased by 15·9 but only by 3·1 ml./min.kg$^{0{\cdot}73}$ after propranolol, while at 10 °C spinal cord thermode temperature oxygen consumption increased by 18·3 but only by 11·2 ml./min.kg$^{0{\cdot}73}$ after propranolol.

(*b*) *Effect of exogenous noradrenaline on the metabolic response to cooling the preoptic area, and the spinal cord.* The results of this series are summarized in Fig. 3. Administration of noradrenaline, 0·5 mg/kg I.M., induced a maximum increase in oxygen consumption of $9{\cdot}1 \pm 1{\cdot}5$ ml./min.kg$^{0{\cdot}73}$, that is, an increase of 48 % above the

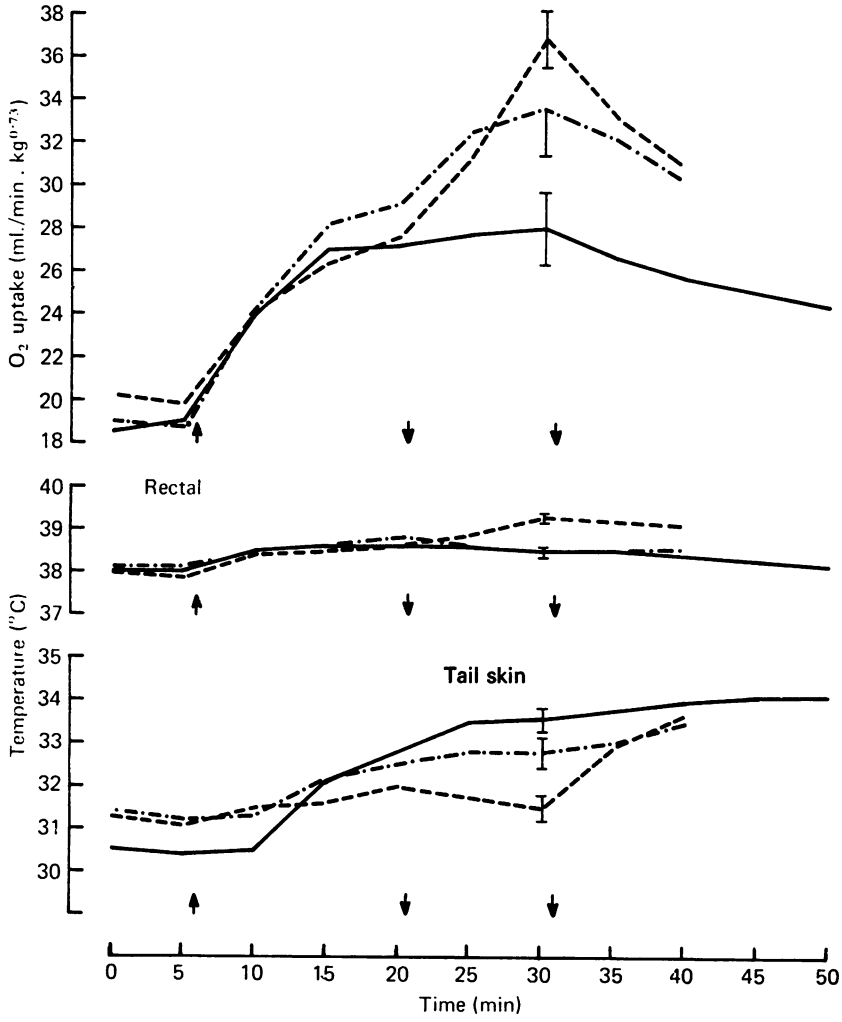

Fig. 3. Effect of noradrenaline (0·5 mg/kg, I.M.) on the response to cooling the preoptic area, and the spinal cord, in seven rats exposed to 24–25 °C. The figure shows the effect of only noradrenaline (continuous line) and that of noradrenaline and cooling the preoptic area (interrupted line) and the spinal cord (dot-interrupted line). The upward pointing arrow shows the injection of noradrenaline and the downward pointing ones show the beginning and the end of thermal stimulation. The vertical lines are the standard errors.

resting metabolic rate. Simultaneous cooling of the preoptic area with water at 10 °C induced, however, an increase of $17{\cdot}9 \pm 1{\cdot}2$, while cooling the spinal cord at the same thermode temperature increased it by $14{\cdot}7 \pm 2{\cdot}0$ ml./min.kg$^{0{\cdot}73}$.

(c) *Effect of warming the preoptic area on the metabolic response to cooling the spinal cord, and vice versa.* The experimental procedure is illustrated in Fig. 4. In this experiment, the spinal cord was cooled for a total of 55 min. When oxygen consumption had stabilized at about 21 ml./min.kg$^{0.73}$, the preoptic area was warmed, which brought the metabolic rate back to the resting level of about 13 ml./min.kg$^{0.73}$, though both core and furred skin temperatures were falling.

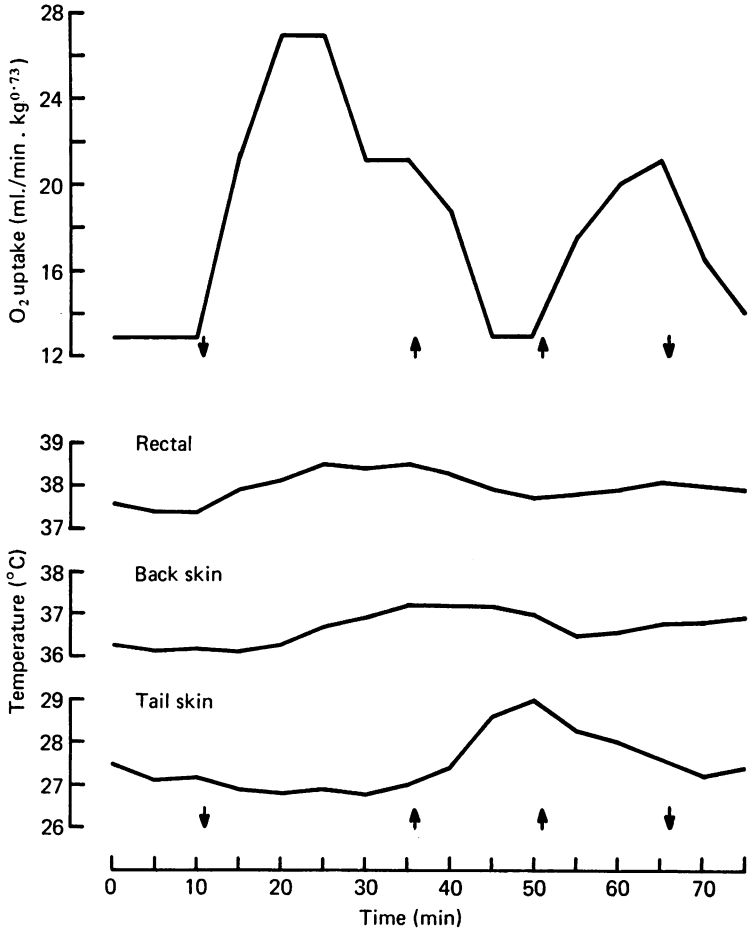

Fig. 4. Effect of cooling the spinal cord and simultaneous warming of the preoptic area in one rat exposed to 24–25 °C. The downward pointing arrows show the beginning and the end of spinal cord cooling, while the upward pointing ones show the beginning and the end of the warm stimulus to the preoptic area.

For further analysis, the average of the last two values before the beginning of thermal stimulation was taken as the resting value. Likewise, the average of the last two values before the other thermoreceptive area was warmed was taken as the value during cold thermal stimulation, and the average of the last two values before the warm stimulation was discontinued was taken as the value during simultaneous cold and warm stimulation, though oxygen consumption in many animals had not yet reached steady state and was still falling. For the post-warming effect, when one of the thermoreceptive areas was still being cooled, the first two values were averaged, though oxygen consumption was still increasing in most animals, and finally, the last two values measured 10–15 min after the cold stimulation was discontinued were averaged and used for the post-stimulation period.

The results of this series are summarized in Fig. 5, where the change in all parameters from their resting level is shown. Cooling the hypothalamus (left side of Fig. 5) induced a significant steady-state increase in oxygen consumption, rectal

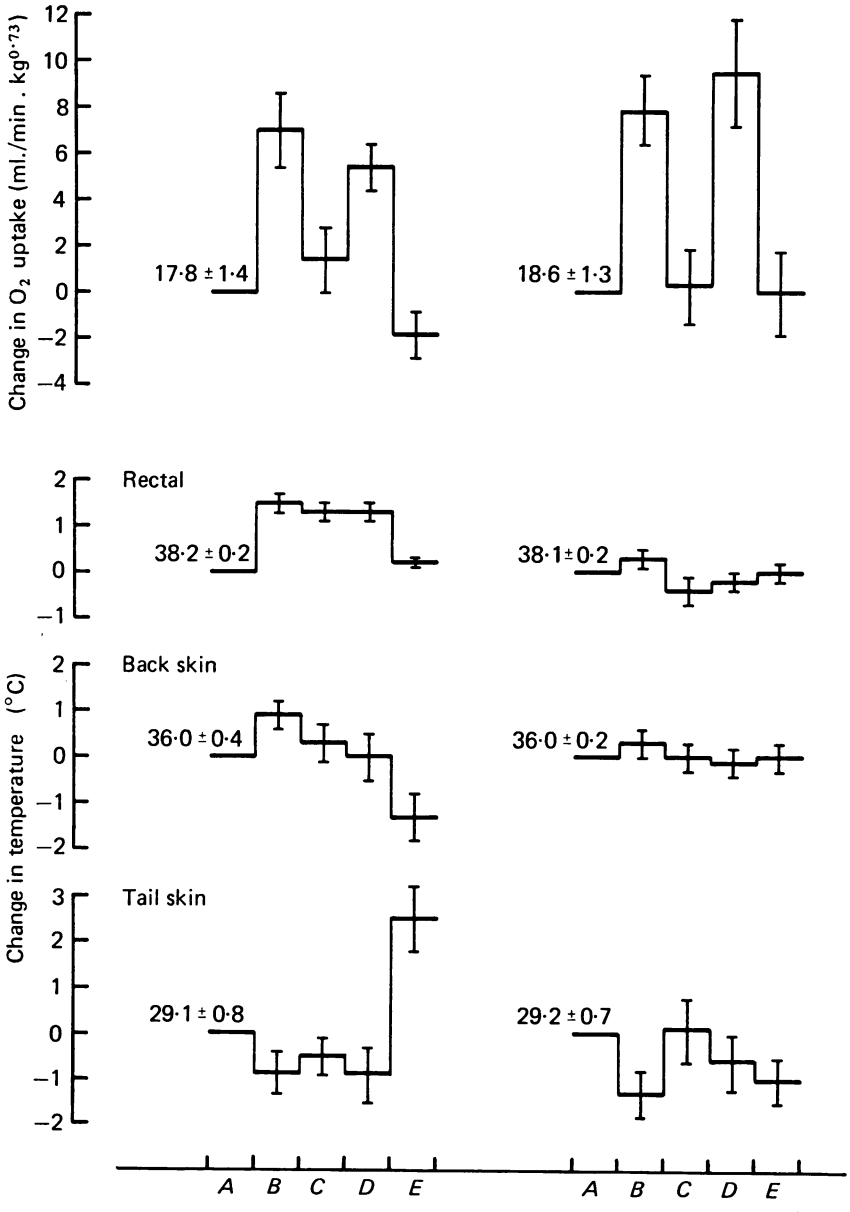

Fig. 5. Effect of warming the spinal cord on the response induced by cooling the preoptic area, and vice versa, in eight rats exposed to 24–25 °C. In *A* are shown the resting values and their standard error. In *B*, *C* and *D* the animals were thermally stimulated by cooling the preoptic area (left side of the Figure) and the spinal cord (right side of the Figure), while in *C* this cold stimulation was counteracted by warming the spinal cord and the preoptic area, respectively; *E* shows the effect 10 min after the thermal stimulation was discontinued. The vertical lines are standard errors.

and back skin temperatures, and a drop in tail skin temperature. Simultaneously warming the spinal cord inhibited the increase in oxygen consumption, though rectal and back skin temperatures fell slightly and tail skin temperature did not significantly differ from its resting level. Cooling the spinal cord (right side of Fig. 5) again induced a significant increase in oxygen consumption and no significant changes in any of the other parameters. Warming the hypothalamus, then, brought the metabolic level back to the resting value, but had no significant effect on any of the other parameters.

DISCUSSION

Three independent series of experiments were done at an ambient temperature which was a compromise between the needs of the various series of experiments. At thermoneutral ambient temperatures, the calorigenic response to exogenous noradrenaline corresponds to the magnitude of non-shivering thermogenesis in the rat (Janský, 1973), but, at this ambient temperature, the metabolic response to cooling the spinal cord (Banet & Hensel, 1976*c*), and probably also the preoptic area, is inhibited by the warm cutaneous thermoreceptors. At ambient temperatures below thermoneutrality, this inhibition ceases and, since skin vessels are already vasoconstricted (Rand, Burton & Ing, 1965), central cold stimulation induces practically no change in unfurred skin temperature (Banet & Hensel, 1976*a*,*c*), which is helpful because skin temperature changes would add an uncontrolled variation in the input to the temperature regulation centre. The ambient temperature used in these experiments (24–25 °C) is, under our experimental conditions, slightly below thermoneutrality (Banet & Hensel, 1977; Banet, Hensel & Pothmann, 1978) and was considered to be a reasonable compromise. That this was so is shown by the fact that cold stimulation of the thermosensitive areas induced significant increases in oxygen consumption but only small changes in tail skin temperature and that, under resting conditions, propranolol decreased the metabolic rate and rectal temperature only slightly, which indicates that the animals were using very little of their capacity for non-shivering thermogenesis (Janský, 1973; Banet & Hensel, 1977).

The first series of experiments shows that the metabolic effects of cooling the preoptic area, and the spinal cord, were directly mediated by the thermosensors in these areas and that propranolol decreased the metabolic response to cooling the spinal cord, but that it did not suppress the metabolic response to cooling the preoptic area. Since propranolol is considered to selectively block non-shivering thermogenesis (see references in the Introduction), one can then conclude that the metabolic increase in propranolol treated animals was induced by shivering. In Fig. 6, the results of the experiments with the two dose levels of propranolol are pooled. The metabolic response to cooling the preoptic area (Fig. 6*A*), and the spinal cord (Fig. 6*B*), represents the total oxygen consumption in each experimental state, while the responses under propranolol represent the oxygen consumption due to shivering and the differences between the two curves represent then the oxygen consumption due to non-shivering thermogenesis. Fig. 6 indicates, therefore, that both thermosensitive areas control both forms of thermogenesis, but that the preoptic area controls mainly non-shivering thermogenesis because only 20 % of

the maximal metabolic response is due to shivering, while the spinal cord controls mainly shivering because only 35 % of the maximal metabolic response is due to non-shivering thermogenesis.

Recent experiments have, however, shown that propranolol interferes with tetanic contraction (Myhre, Böed & Aars, 1977) and the possibility that it could inhibit shivering cannot be disregarded. If propranolol does inhibit shivering, our conclusion that the spinal cord controls non-shivering thermogenesis could be wrong

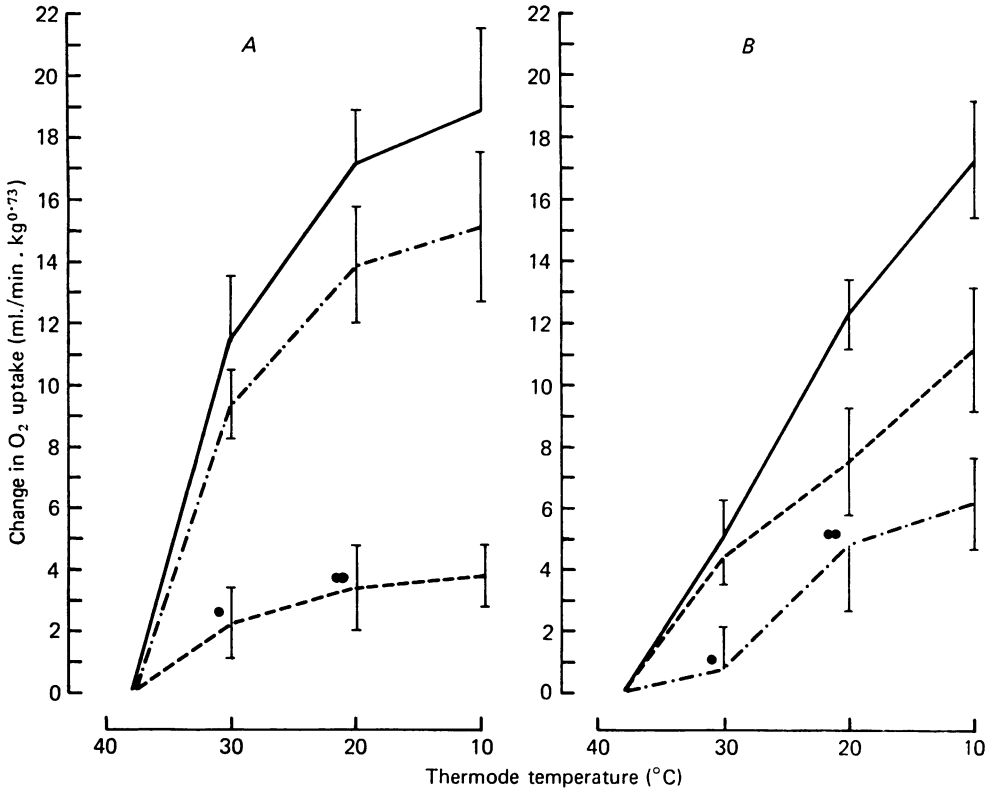

Fig. 6. Average changes in oxygen consumption induced by cooling the preoptic area (left panel *A*) and the spinal cord (right panel *B*) in nine rats exposed to 24–25 °C during infusion of saline (continuous line) and propranolol (interrupted line). The third line (dot-interrupted) shows the difference between the other two and is considered to represent the fraction of oxygen consumption due to non-shivering thermogenesis. All changes are highly significant ($P < 0{\cdot}005$) except those marked with two dots, which are probably significant ($P < 0{\cdot}05$), and those with one dot that are not significant. The vertical lines are standard errors.

because the fall in metabolism induced by propranolol when this area is cooled might have been due only to the inhibition of shivering. On the other hand, our conclusion that the preoptic area controls both forms of heat production would still be valid, but the contribution of non-shivering thermogenesis to the total metabolic response would then be less than the estimate in Fig. 6*A* because the fall in metabolism would have been partially due to the inhibition of shivering rather than to the inhibition

of only non-shivering thermogenesis. In other words, propranolol would overestimate the capacity for non-shivering thermogenesis.

The second series of experiments shows that the animals had a capacity for non-shivering thermogenesis, as estimated by exogenous noradrenaline, of $9{\cdot}1 \pm 1{\cdot}5$ ml. O_2/min.kg$^{0{\cdot}73}$, that is, 48 % of the resting metabolic rate. This is a relatively high capacity for rats adapted to thermoneutrality (Janský, 1973; Banet & Hensel, 1976*a*,*b*), but it is significantly lower than the $15{\cdot}1 \pm 2{\cdot}3$ ml. O_2/min.kg$^{0{\cdot}73}$ induced by perfusing the preoptic thermode with water at 10 °C and estimated by administration of propranolol (Fig. 6*A*). On the assumption that propranolol does not significantly interfere with shivering, it can be concluded that noradrenaline underestimates the capacity for non-shivering thermogenesis, either because it does not stimulate all adrenergic receptors controlling non-shivering thermogenesis or because there are other forms of non-shivering thermogenesis that are not stimulated by noradrenaline but are blocked by propranolol.

The calorigenic response to noradrenaline and simultaneous cooling of the preoptic area was $17{\cdot}9 \pm 1{\cdot}2$ ml. O_2/min.kg$^{0{\cdot}73}$ (Fig. 3), that is, 2·8 and 8·8 ml. O_2/min.kg$^{0{\cdot}73}$ higher than the capacity for non-shivering thermogenesis as estimated with propranolol and noradrenaline, respectively. This means that at least 2·8 ml. O_2/min.kg$^{0{\cdot}73}$ was due to shivering, amount which is rather close to the propranolol estimate of shivering in Fig. 6*A*. The calorigenic response to noradrenaline and simultaneous cooling of the spinal cord was $14{\cdot}7 \pm 2{\cdot}0$ ml. O_2/min.kg$^{0{\cdot}73}$ (Fig. 3). If the metabolic response induced by cooling the spinal cord were due only to shivering, a significantly higher response could have been expected because the metabolic increase induced by cooling this area ($17{\cdot}2 \pm 1{\cdot}9$ ml. O_2/min.kg$^{0{\cdot}73}$, Fig. 6*B*) would have added to that induced by noradrenaline (9·1 ml. O_2/min.kg$^{0{\cdot}73}$, Fig. 3). On the other hand, this low metabolic response may indicate that the thermal stimulation of the spinal cord was not as effective as in the previous series because the warm blood draining from the cervical adipose tissue, stimulated by noradrenaline, counteracted the cooling effect of the thermode.

The last series of experiments shows that the metabolic effect of cooling one of the thermosensitive areas can be fully counteracted by warming the other thermosensitive area and that this effect was, furthermore, mediated by the central thermosensors themselves because the changes in neither core nor skin temperature can explain it (Fig. 5). It has been reported (Fuller *et al.* 1975, 1977) that in rats exposed to cold, warming the preoptic area increased shivering, while warming the spinal cord increased non-shivering thermogenesis. Our present series shows, however, that warming the preoptic area inhibited the metabolic increase induced by cooling the spinal cord to a temperature (30 °C) that induced only shivering (Fig. 6*B*), while warming the spinal cord inhibited the metabolic increase induced by cooling the preoptic area to 30 °C, that is, it inhibited non-shivering thermogenesis (Fig. 6*A*). The difference between our conclusions and those of Fuller and co-workers may be due to the fact that we used warmer thermode temperatures; on the other hand, the quantitative estimate of shivering made by these authors was based on the frequency of shivering bursts and two important parameters, the amplitude and duration of the bursts (Hemingway, 1963), were not considered.

Taken all together, the three series of experiments clearly show that the

thermosensors in the preoptic area control both thermogenic mechanisms, that the warm sensors in the spinal cord control non-shivering thermogenesis and that the cold ones control shivering; the evidence that these cold sensors control also non-shivering thermogenesis is, however, not conclusive because of the possibility that propranolol might inhibit shivering. It has, however, been suggested that a significant amount of the metabolic increase induced by cooling the spinal cord is due to non-shivering thermogenesis (Banet & Hensel, 1976*b*,*c*). Prolonged and repetitive cooling of this area, furthermore, induced an increase in the capacity for non-shivering thermogenesis (Banet & Hensel, 1976*b*) which was most likely mediated by the sympathetic nervous system because thyroxine, a hormone that might have mediated this increase in the capacity for non-shivering thermogenesis (LeBlanc & Villemaire, 1970), appears not to be stimulated by cooling the spinal cord of the pig (Evans & Ingram, 1974). It would then appear logical to conclude that the cold thermosensors in the spinal cord control also non-shivering thermogenesis and that, therefore, the estimated contributions of shivering and non-shivering thermogenesis to the total metabolic response in Fig. 6 are essentially correct.

The above discussion shows that the working hypothesis first developed for the guinea-pig (Brück & Wünnenberg, 1970), that the spinal cord controls essentially only shivering and the preoptic area only non-shivering thermogenesis, is valid only at low intensities of cold stimulation since at higher intensities of stimulation both thermoreceptive areas elicit both forms of heat production. This is so because, as Fig. 6 shows, both forms of thermogenesis do not seem to have the same threshold. Thus, the preoptic threshold for non-shivering thermogenesis seems to be lower than that of shivering, while the spinal cord threshold for shivering seems to be lower than that of non-shivering thermogenesis and, therefore, only strong cooling will elicit the thermogenic form with higher threshold. If this hypothesis is correct, the quantitative contribution of the thermogenic form with higher threshold should increase as the stimulating temperature decreases. That this is so is suggested by the increase in non-shivering thermogenesis induced by cooling the spinal cord (Fig. 6*B*), increase that is inversely proportional to thermode temperature and that does not seem to have reached a maximum at the lowest thermode temperatures used in these experiments. The metabolic increase induced by cooling the preoptic area (Fig. 6*A*) suggests, however, that it has more or less reached its maximum and that, therefore, lower thermode temperatures are not likely to increase the intensity of shivering, but this is most probably due to the fact that, contrary to cooling the spinal cord, preoptic cooling induces a significant increase in body temperature (Fig. 2) which counteracts the effect of local cooling and, therefore, limits its effectiveness. Thus, our hypothesis, which suggests that the hypothalamic thermosensors still play a role in animals who lost their capacity for non-shivering thermogenesis due to aging or adaptation to heat, needs further research but appears at present to be tenable.

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REFERENCES

ALEXANDER, G. & WILLIAMS, D. (1968). Shivering and non-shivering thermogenesis during summit metabolism in young lambs. *J. Physiol.* **198**, 251–276.

BANET, M. & HENSEL, H. (1976*a*). Nonshivering thermogenesis induced by repetitive hypothalamic cooling in the rat. *Am. J. Physiol.* **230**, 522–526.

BANET, M. & HENSEL, H. (1976*b*). Nonshivering thermogenesis induced by repetitive cooling of spinal cord in the rat. *Am. J. Physiol.* **230**, 720–723.

BANET, M. & HENSEL, H. (1976*c*). The interaction between cutaneous and spinal thermal inputs in the control of oxygen consumption in the rat. *J. Physiol.* **260**, 461–473.

BANET, M. & HENSEL, H. (1977). The control of shivering and non-shivering thermogenesis in the rat. *J. Physiol.* **269**, 669–676.

BANET, M., HENSEL, H. & POTHMANN, B. (1978). Autonomic thermoregulation after intermittent cooling of the spinal cord and cold exposure in the rat. *J. Physiol.* **275**, 439–447.

BANET, M. & SEGUÍN, J. J. (1967). A thermode for rat brain. *Electroenceph. clin. Neurophysiol.* **23**, 572–573.

BLATTEIS, C. M. (1976). Effect of propranolol on endotoxin-induced pyrogenesis in newborn and adult guinea pigs. *J. appl. Physiol.* **40**, 35–39.

BRODY, S. (1945). *Bioenergetics and Growth.* New York: Reinhold.

BRÜCK, K. & WÜNNENBERG, B. (1965). Blockade der chemischen Thermogenese und Auslösung von Muskelzittern durch Adrenolytica und Ganglienblockade beim neugeborenen Meerschweinchen. *Pflügers arch. ges. Physiol.* **282**, 376–389.

BRÜCK, K. & WÜNNENBERG, W. (1970). 'Meshed' control of two effector systems: nonshivering and shivering thermogenesis. In *Physiological and Behavioral Temperature Regulation*, ed. HARDY, J. D., GAGGE, A. P. & STOLWIJK, J. A. J., pp. 562–580. Springfield, Ill.: Thomas.

EVANS, S. E. & INGRAM, D. L. (1974). The significance of deep body temperature in regulating the concentration of thyroxine in the plasma of the pig. *J. Physiol.* **236**, 159–170.

FULLER, C. A., HOROWITZ, J. M. & HORWITZ, B. A. (1977). Spinal cord thermosensitivity and sorting of neural signals in cold-exposed rats. *J. appl. Physiol.* **42**, 154–158.

FULLER, C. A., HORWITZ, B. A. & HOROWITZ, J. M. (1975). Shivering and nonshivering thermogenesis responses of cold-exposed rats to hypothalamic warming. *Am. J. Physiol.* **228**, 1519–1524.

HART, J. S. (1971). Rodens. In *Comparative Physiology of Thermoregulation*, vol. II, *Mammals*, ed. WHITTOW, G. C., pp. 1–149. New York, London: Academic.

HEIM, T. & HULL, D. (1966). The effect of propranolol on the calorigenic response in brown adipose tissue in new-born rabbits to catecholamines, glucagon, corticotrophin and cold exposure. *J. Physiol.* **187**, 271–283.

HEMINGWAY, A. (1963). Shivering. *Physiol. Rev.* **43**, 397–422.

HORWITZ, B. A. & HANES, G. E. (1976). Propranolol and pyrogen effects on shivering and nonshivering thermogenesis in rats. *Am. J. Physiol.* **230**, 637–642.

HSIEH, A. C. L. & CARLSON, L. D. (1957). Role of adrenaline and noradrenaline in chemical regulation of heat production. *Am. J. Physiol.* **190**, 243–246.

HSIEH, A. C. L., CARLSON, L. D. & GRAY, G. (1957). Role of the sympathetic nervous system in the control of chemical regulation of heat production. *Am. J. Physiol.* **190**, 247–251.

JANSKÝ, L. (1973). Non-shivering thermogenesis and its thermoregulatory significance. *Biol. Rev.* **48**, 85–132.

LEBLANC, J. A. & VILLEMAIRE, A. (1970). Thyroxine and noradrenaline on noradrenaline sensitivity, cold resistance, and brown fat. *Am. J. Physiol.* **218**, 1742–1745.

MURGATROYD, D. & HARDY, J. D. (1970). Central and peripheral temperatures in behavioral thermoregulation of the rat. In *Physiological and Behavioral Temperature Regulation*, ed. HARDY, J. D., GAGGE, A. P. & STOLWIJK, J. A. J., pp. 874–891. Springfield, Ill.: Thomas.

MYHRE, L., RÖED, A. & AARS, H. (1977). Inhibitory effect of propranolol on tetanic contraction in rabbit. *Eur. J. Pharmac.* **42**, 355–361.

RAND, R. P., BURTON, A. C. & ING, T. (1965). The tail of the rat, in temperature regulation and acclimatization. *Can. J. Physiol. Pharmacol.* **43**, 257–267.

REICHLIN, S. (1964). Function of the hypothalamus in regulation of pituitary-thyroid activity. In *Brain Thyroid Relationship*, ed. CAMERON, M. P. & O'CONNOR, M., pp. 17–34. Boston: Little Brown.

SATINOFF, E. (1964). Behavioral thermoregulation in response to local cooling of the rat brain. *Am. J. Physiol.* **206**, 1389–1394.

SCHÖNBAUM, E., JOHNSON, G. E., SELLERS, E. A. & GILL, M. J. (1966). Adrenergic β-receptors and non-shivering thermogenesis. *Nature, Lond.* **210**, 426.

STONER, H. B. (1972). Effect of injury on the responses to thermal stimulation of the hypothalamus. *J. appl. Physiol.* **33**, 665–671.