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DECEREBRATION ACTIVATES THERMOGENESIS IN THE RAT

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SUMMARY

1. Under fluothane anaesthesia, suction decerebration was performed at the immediate pre-pontine level in adult, male, Sprague–Dawley rats; this resulted in a large and sustained rise in rectal temperature from 35·6 ± 0·2 (control) to 38·8 ± 0·5 °C (decerebrate) following recovery from anaesthesia. Propranolol inhibited this rise.

2. In a separate group of continuously (urethane) anaesthetized rats, brain transection at the immediate pre-pontine level produced marked increases in rectal temperature and oxygen consumption, both of which were inhibited by injection of the β-adrenergic antagonist propranolol (10 mg/kg).

3. The rise in rectal temperature (2·8 ± 0·4 °C) after transection was preceded by a greater increase (3·6 ± 0·3 °C) in the temperature of the interscapular brown adipose tissue (i.b.a.t.). Skin temperature on the tail showed no immediate response.

4. In anaesthetized lean (+/?) male Zucker rats, rectal and i.b.a.t. temperatures showed similar responses to Sprague–Dawley rats after decerebration, but in the genetically obese (fa/fa) Zucker rat, temperatures were not significantly altered by decerebration.

5. The above results, together with macroscopic examination of the transected brains, suggest that descending pathways (possibly arising in the mid-brain tegmentum) normally inhibit a sustained thermogenic drive from areas in the lower brain stem. Decerebration can release this inhibition and cause a large rise in body temperature and in metabolic rate, which apparently result from sympathetic activation of i.b.a.t. The genetically obese Zucker rat exhibits an impaired thermogenic response to decerebration.

INTRODUCTION

Environmental temperature and food intake are two of the major factors influencing metabolic rate in mammals. Chronic exposure to cold and high levels of food intake both cause increases in energy expenditure, known as non-shivering thermogenesis (n.s.t.) and diet-induced thermogenesis (d.i.t.) respectively, whereas high ambient temperatures or reductions in food intake cause depression of metabolic rate. Studies on laboratory rodents have demonstrated that n.s.t. and d.i.t. result from sympathetic activation of brown adipose tissue (Foster & Frydman, 1978; Rothwell & Stock, 1981).

The involvement of the hypothalamus in thermoregulation has been clearly

established from pharmacological and neurophysiological studies, with the pre-optic and anterior areas playing a major role (see Gale, 1973; Hensel, 1973 for reviews). The ventromedial and lateral areas of the hypothalamus are known to be involved in the regulation of body weight, but it has been assumed that this is achieved by effects on food intake (see Novin, Wyrwicka & Bray 1976; Kissileff & Van Itallie, 1982 for reviews). However, it has recently been demonstrated that electrical stimulation of the ventromedial hypothalamus causes activation of brown adipose tissue (Perkins, Rothwell, Stock & Stone, 1981), whereas electrolytic destruction of this area results in atrophy and reduced activity of brown fat (Seydoux, Rohner-Jeanrenaud, Assimacopoulos-Jeannet, Jeanrenaud & Girardier, 1981). These results, together with observations of increased metabolic efficiency (i.e. reduced d.i.t.) in animals with ventromedial hypothalamic lesions (Han, 1967; Goldman, Bernardis & Frohman, 1974), also implicate this area in the control of thermogenesis. In contrast, Bignall, Heggenness & Palmer (1975) have reported that temperature regulation is only partially impaired in neonatal rats after surgical removal of the hypothalamus. This group has also shown that decerebration of neonatal rats produces maximal thermogenesis and a large increase in rectal temperature, both of which are unaffected by fasting or by high ambient temperature. These findings suggest that thermogenesis is normally controlled by descending inhibition of centres, probably located in the brain stem, which in turn control heat generating systems. Bignall *et al.* (1975) assumed that the increased heat production following decerebration was due to brown adipose tissue thermogenesis, but this assumption was not tested and studies were not performed on adult animals because the authors claimed that changes in muscle tension and hyperactivity followed decerebration.

We have investigated this problem to see if an elevation of metabolic rate occurs in decerebrated adult rats and whether this involves sympathetic activity and activation of brown adipose tissue. The effects of decerebration on the activity of brown adipose tissue in genetically obese (fa/fa) Zucker rats, which have been reported to exhibit defective thermogenic responses to cold (Trayhurn & James, 1981) and diet (Young, Tulp & Horton, 1980; Rothwell, Saville & Stock, 1981), were also studied.

METHODS

In the first experiment, five adult (3 month) male Sprague–Dawley SPF rats weighing approximately 350 g were anaesthetized with fluothane and a suction decerebration was performed at the immediate pre-pontine level (Fig. 1). Five control animals were anaesthetized and allowed to recover without receiving any surgical treatment. All animals were allowed to recover from the anaesthetic in a room at 24–25 °C and rectal temperature was recorded over the next 3 h. In a later extension to this experiment, four decerebrate rats were injected with propranolol (10 mg/kg, I.P.) after recovery from anaesthesia.

Separate groups of Sprague–Dawley rats, similar to those described above, were anaesthetized with urethane (0·15/100 g, I.P.) and the brain was transected by insertion of a cauterizing lance at the pre-pontine level ($n = 7$). Control animals were anaesthetized and received either no transection ($n = 5$) or a transection 1–4 mm anterior to the pre-pontine level ($n = 5$); the latter were classified as pre-collicular sections (see Fig. 1). After a 1 h recovery period at room temperature (24–25 °C), oxygen consumption ($\dot{V}_{O_2}$) was measured in a closed circuit respirometer (Stock, 1975) at thermoneutrality (29 °C) for 2 h before and after injection of propranolol (10 mg/kg, I.P.). Values were corrected for body size (ml/min . kg $(W)^{0.75}$) and all rats were maintained under anaesthesia

throughout the experiment. Rectal temperatures were recorded at the end of each 2 h $\dot{V}_{O_2}$ measurement.

In the third study, Sprague–Dawley rats ($n = 4$) were anaesthetized with urethane and a thermocouple (Comark, Sussex) placed under the interscapular brown adipose tissue (i.b.a.t.) depot; the wound was closed with surgical clips. A similar plastic-coated thermocouple was inserted 5 cm into the rectum. I.b.a.t. and rectal temperatures were recorded continuously for approximately 1 h before and up to 7 h after transection of the brain at the immediate pre-pontine level. Skin temperature was recorded in two animals from an extra thermocouple placed on the tail approximately 5 cm from the body.

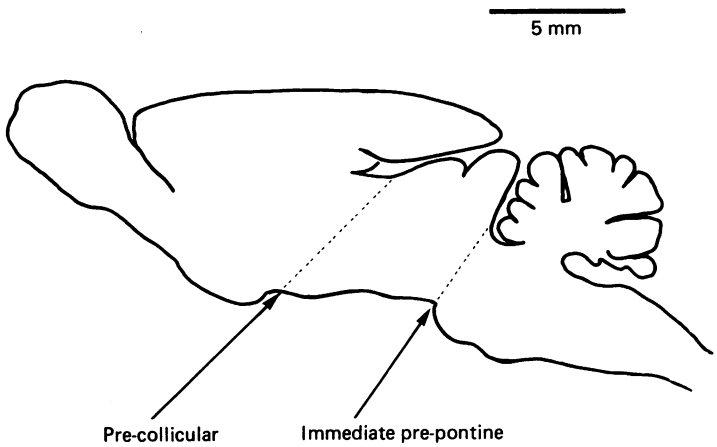

Fig. 1. Outline of rat brain (2 mm lateral to the mid line) showing the levels of section defined as pre-collicular i.e. from the rostral edge of the superior colliculus dorsally to the optic tract ventrally, and immediate pre-pontine i.e. from the caudal edge of the inferior colliculus dorsally to the rostral limit of the pons ventrally. Although, when the levels of decerebration were classified from the frozen brain sections, the plane of decerebration frequently differed by 1 or 2 mm from the ideal planes shown, most decerebrations clearly fell into one of two groups. The criterion adopted for the occasional intermediate level of section was that if, at any point, not more than 1 mm of tissue was present rostral to the plane shown, the decerebration was classified as immediate pre-pontine; if more than 1 mm of tissue was present, the decerebration was included in the pre-collicular group. Decerebrations caudal to the immediate pre-pontine plane were included in the immediate pre-pontine group.

Rectal and i.b.a.t. temperatures were also recorded in urethane anaesthetized adult (4–6 month) male, lean (homozygotic or heterozygotic, +/?) and obese (homozygotic, fa/fa) Zucker rats (average weight 350 and 550 g respectively) both before and after transection of the brain. Skin temperature was also recorded from the tail in three animals.

The level of decerebration after transection was later confirmed by macroscopic examination of frozen longitudinal sections of the brain (Fig. 1). All values for $\dot{V}_{O_2}$ and temperature have been expressed as means $\pm$S.E. of the mean. Statistical comparisons were made using Student's t test for matched and unmatched data, as appropriate; the quoted probabilities are two-tailed.

RESULTS

In the first experiment, on animals subsequently allowed to recover from the anaesthetic, suction decerebration caused rectal temperature to rise to 38·8 $\pm$ 0·5 °C over the 3 h period following the operation, whereas in control animals, rectal

temperature remained relatively constant (35·6 ± 0·2, $P < 0.001$). Rectal temperature also remained constant (35·0 ± 0·6 °C) in the four decerebrate rats injected with propranolol. However, in some animals, suction decerebration caused fairly severe blood loss and in two experimental rats muscular spasms occurred. In these two cases the spasms were followed by death, 4 h after the operation, associated with rectal temperatures of 40·1 and 40·2 °C respectively. Nevertheless, in experimental rats, and those receiving propranolol, there was no constant association between locomotor activity, elevated muscle tone or muscle spasms on the one hand, and body temperature on the other.

TABLE 1. Oxygen consumption ($\dot{V}_{O_2}$) and rectal temperature of Sprague–Dawley rats

	Intact C.N.S.	Pre-collicular transection	Immediate pre-pontine transection
$\dot{V}_{O_2}$ (ml/min . $W^{0.75}$)			
Before propranolol	10·49 ± 0·10	9·27 ± 0·91	18·88 ± 0·60***
After propranolol	10·16 ± 0·42	8·64 ± 0·54	11·93 ± 0·41*
Rectal temperature (°C)			
Before propranolol	36·2 ± 0·05	35·2 ± 0·6	39·2 ± 0·4***
After propranolol	34·9 ± 0·09	34·1 ± 0·4	37·3 ± 0·35*
	$n = 5$	$n = 5$	$n = 7$

* $P < 0.05$, *** $P < 0.001$ compared to controls.

The effects of brain transection in anaesthetized Sprague–Dawley rats upon $\dot{V}_{O_2}$ and rectal temperature are shown in Table 1. $\dot{V}_{O_2}$ and rectal temperature were slightly lower in rats transected at the pre-collicular level (Fig. 1) than in those which were not transected. However, transection at the pre-pontine level produced large increases in $\dot{V}_{O_2}$ and body temperature which were sustained over the 2 h of the measurements. Injection of propranolol had no significant effect on $\dot{V}_{O_2}$ in control rats or those with the anterior level of transection, although rectal temperature was reduced. However, following the administration of propranolol, either intravenously (2 mg/kg) or I.P. (10 mg/kg), the elevated $\dot{V}_{O_2}$ of animals with the immediate pre-pontine transections was markedly reduced, to a value only slightly above that of controls. In two additional animals with immediate pre-pontine transactions, injection of atropine (2 mg/kg, I.P.) did not alter the temperature response.

In anaesthetized Sprague–Dawley rats, i.b.a.t. temperature was slightly (0·5–1·0 °C) below rectal before decerebration, but transection produced a rapid rise beginning 30–60 s after decerebration in most animals, although some rats showed an initial small reduction in temperature (see Fig. 2). However, in all cases the temperature of the interscapular brown fat rose before the rectal and achieved a higher peak value (absolute rise, i.b.a.t. 3·6 ± 0·3, rectal 2·8 ± 0·4 °C). The rate of increase of i.b.a.t. and rectal temperatures was rather variable; subjectively it appeared rather slower in those animals in which the amount of blood loss during decerebration was above average. Nevertheless, a steady peak temperature was achieved in all rats within 40 min of decerebration. Immediately after decerebration, tail temperature did not normally change significantly, although subsequently it slowly increased.

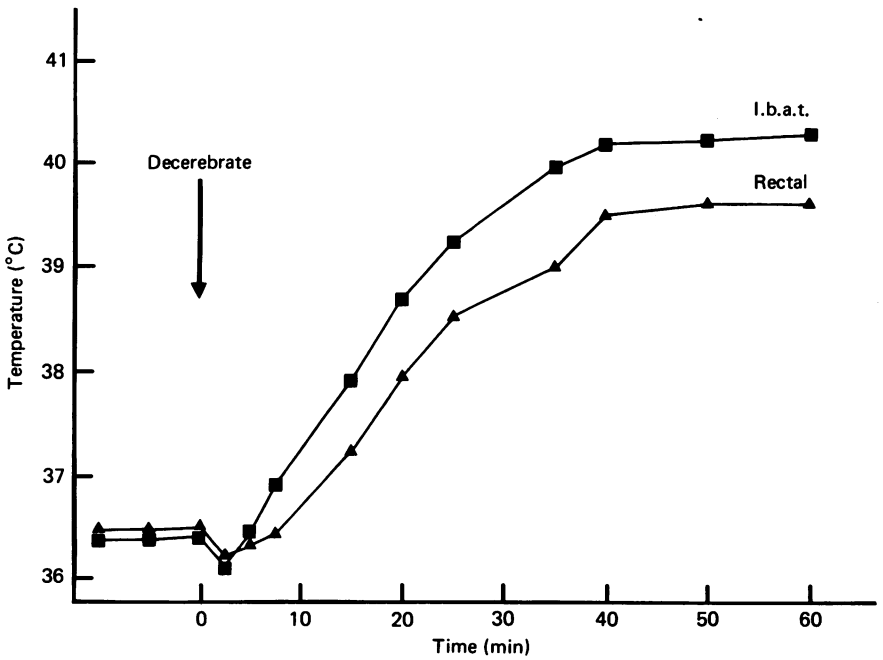

Fig. 2. Rectal and i.b.a.t. temperatures in an anaesthetized Sprague–Dawley rat, before and after decerebration at the immediate pre-pontine level (transection at time 0).

TABLE 2. Rectal and i.b.a.t. temperatures of lean and obese Zucker rats before and after decerebration

	Lean	Obese
Rectal temperature (°C)		
Before decerebration	35·4 ± 0·2	35·8 ± 0·35
After decerebration	38·0 ± 0·4***	34·5 ± 0·4
I.b.a.t. temperature (°C)		
Before decerebration	35·4 ± 0·1	35·6 ± 0·4
After decerebration	38·7 ± 0·4***	35·4 ± 0·5
	$n = 6$	$n = 9$

*** $P < 0·001$ compared to temperature before decerebration.

The temperature responses to decerebration in lean Zucker rats (Table 2) were similar to those of Sprague–Dawley rats, but were less variable. Before decerebration, rectal and i.b.a.t. temperatures were almost identical, but pre-pontine transection produced a rapid increase in i.b.a.t. temperature (30 s latency, rate 0·08 ± 0·02 °C/min) followed by a sustained rise to the plateau value. The latency of the rectal temperature rise was 2–3 min and both the rate of increase and the maximum value achieved were lower than for i.b.a.t. Average i.b.a.t. and rectal temperatures of genetically obese Zucker rats were slightly reduced after transection (Table 2), but three out of the nine animals did show increases in i.b.a.t. temperature (0·6, 1·5 and 2·0 °C), whereas temperature fell by up to 2 °C in the other six animals.

Macroscopic examination of thick frozen sections of the brains revealed that all rises in temperature and $\dot{V}_{O_2}$ were associated with immediate pre-pontine decerebrations whereas little or no such effects were produced with transections at the pre-collicular level. Preliminary experiments with deep transverse mid-line cuts (approximately 2 mm wide) at the level of the anterior pons produced the same response as a complete transection, but shallow cuts (less than about 5 mm below the cortical surface) were without such effects.

DISCUSSION

The initial experiment, on adult rats allowed to recover after suction decerebration, supports the findings of Bignall *et al.* (1975) in neonatal rats. Decerebration, carried out at the immediate pre-pontine level under direct vision, produced a large and sustained rise in rectal temperature which did not appear to be associated with increased physical activity immediately after the operation. However, some animals did develop muscular spasms after a few hours; it is possible that these spasms together with the rather variable and sometimes severe blood loss, could have influenced the results, although marked temperature rises did occur in the absence of any obviously increased muscle activity. Furthermore, the fact that β-adrenergic blockade with propranolol inhibited the rise in rectal temperature would suggest that the hyperthermic response to decerebration was mediated by catecholamine release.

Transection of the brain with a cauterizing lance caused very little blood loss and, in these animals maintained under continuous urethane anaesthesia, there was no overt physical activity and muscle tone was virtually absent. Animals transected at the pre-collicular level showed slightly lower rectal temperatures and $\dot{V}_{O_2}$ than controls, but transection at the immediate pre-pontine level caused a large increase in temperature and metabolic rate, both of which were inhibited by the β-adrenergic antagonist propranolol. This suggests that the rise in temperature was due to an increase in metabolic rate mediated by the sympathetic nervous system. Propranolol had little or no effect on $\dot{V}_{O_2}$ in control rats so that the fall in rectal temperature in these animals was presumably due to increased heat loss.

β-adrenergic blockade also inhibits the high metabolic rates of both cold-adapted animals exhibiting n.s.t. and over-fed rats exhibiting d.i.t., but it has virtually no effect on normally fed animals housed in a warm environment (Rothwell & Stock, 1979). The major effector tissue for n.s.t. and d.i.t. is brown fat, which is also mainly responsible for the thermogenic response to exogenous noradrenaline (Foster & Frydman, 1978; Rothwell & Stock, 1981). The results obtained in the third and fourth experiments, on anaesthetized Sprague–Dawley and lean Zucker rats, suggest that the rise in metabolic rate after decerebration also results from thermogenesis in i.b.a.t. The dramatic temperature rise of i.b.a.t. (+3 °C) after transection is very much greater than that observed after either intravenous injection of noradrenaline or electrical stimulation of the ventromedial hypothalamus – i.e. a rise of about 1 °C (Perkins *et al.* 1981). It is unlikely to have resulted from passive warming of the tissue since core temperature rose more slowly than i.b.a.t. and achieved a lower peak value. The absence of any immediate change in tail temperature suggests that the transections did not cause peripheral vasoconstriction. However, the initial small

decrease in i.b.a.t. temperature seen in some Sprague–Dawley rats is similar to that observed after either rapid intravenous injection of noradrenaline, stimulation of the sympathetic nerves directly supplying the i.b.a.t. depot (Flaim, Horwitz & Horowitz, 1976) or stimulation of the ventromedial hypothalamus (Perkins *et al.* 1981). This initial small reduction has previously been ascribed to a transient vasoconstriction. The rapid rise in i.b.a.t. temperature after decerebration could always be inhibited by injection of propranolol.

Lean Zucker rats showed very similar temperature responses to Sprague–Dawley rats, with immediate pre-pontine brain transection producing a large and rapid increase in i.b.a.t. temperature and a delayed rise in rectal temperature. However in the case of the nine obese Zucker rats, average i.b.a.t. and rectal temperature declined slightly after decerebration. Although these mutants are noted for their hyperphagia, they will still deposit excess fat when their intake is restricted to the level of their lean litter-mates, suggesting that the major metabolic lesion in this animal is a high energetic efficiency, i.e. a reduced energy expenditure (Young *et al.* 1980; Rothwell *et al.* 1981). Obese Zucker rats, like genetically obese (ob/ob) mice, show defective n.s.t. (Trayhurn & James, 1981) and d.i.t. (Young *et al.* 1980) but can respond normally to exogenous noradrenaline (Rothwell *et al.* 1981). This suggests that peripheral thermogenic mechanisms can function normally when given an adequate stimulus and that the low thermogenic responses to food and cold may be due to a nervous system defect. The findings of this study suggest that any such thermogenic defect in the obese Zucker rat must be located either in the peripheral nervous system or in the lower brain stem. The blunted metabolic response to food in this mutant can be enhanced by injection of atropine sulphate (Rothwell *et al.* 1981), but atropine did not alter the temperature responses to decerebration.

Macroscopic examination of brain slices indicated that maximal thermogenic responses were achieved with transections at the immediate pre-pontine level, transections 1–4 mm anterior to this level being without effect, as were shallow cuts (less than about 5 mm below the brain surface). When a rise in temperature had been established after decerebration, it was not inhibited or potentiated by further, more caudal pontine transections, although it is known that transections of the upper cervical spinal cord do suppress i.b.a.t. thermogenesis (Smith & Horwitz, 1969).

Thus, it seems that descending pathways, possibly arising in the mid-brain tegmentum inhibit the thermogenic drive arising in areas of the lower brain stem. Immediate pre-pontine decerebration releases this inhibition and causes a large rise in body temperature and metabolic rate, the latter apparently resulting from sympathetic activation of brown adipose tissue.

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