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Thyrotropin releasing hormone-induced hyperthermia in mice: possible involvement of adrenal and pituitary glands

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1 To investigate the hyperthermic effect of thyrotropin releasing hormone (TRH) and its potentiation by exogenous catecholamines (CA), the role of the adrenal medulla and of the pituitary gland was studied in unoperated, adrenal-demedullated or hypophysectomized mice.

2 In unoperated mice, TRH 40 mg kg^{-1} i.p. produced a hyperthermia which was accompanied by an increase in plasma noradrenaline (NA) and adrenaline (Ad). NA or Ad, both at a dose of 1 mg kg^{-1} i.p., enhanced the TRH-induced hyperthermia.

3 Adrenal demedullation suppressed the hyperthermia and the increase of plasma CA produced by TRH but not the potentiation of this hyperthermia by exogenous CA.

4 Hypophysectomy abolished the TRH-induced hyperthermia but not the increase of plasma CA or the potentiation of this hyperthermia by exogenous CA.

5 These results suggest that, in mice, both the adrenal medulla and the pituitary gland play an essential role in TRH-induced hyperthermia but not in its potentiation.

Introduction

Thyrotropin releasing hormone (TRH) is one of the neuropeptides which is capable of modifying thermoregulation. Early studies demonstrated that TRH given intracerebroventricularly (i.c.v.) caused a marked hypothermia in cats (Metcalf, 1974). On the other hand, hyperthermia was observed when TRH was injected i.c.v. in rabbits (Horita & Carino, 1975) and in rats (Prasad, Matsui, Williams & Peterkofsky, 1978; Boschi & Rips, 1981a) or into the preoptic area of the hypothalamus in rats (Boschi & Rips, 1981a). However, in mice, i.c.v. injection of TRH had no effect on rectal temperature (Boschi & Rips, 1981b). Peripheral injection of TRH induced hyperthermia which was potentiated by α-adrenoceptor agonists and inhibited by α-adrenoceptor antagonists (Desiles & Rips, 1981). Noradrenaline (NA) given intraperitoneally (i.p.) strongly potentiated TRH-induced hyperthermia whereas NA given i.c.v. did not (Boschi & Rips, 1981b). These findings have demonstrated that the TRH-induced hyperthermia and its potentiation involved an α-adrenergic system. However, the complete mechanism through which TRH produces these hyperthermic responses is still uncertain.

The adrenal medulla releases catecholamines (CA) and the pituitary gland secretes peptides (Jansky, 1973; Huidobro-Toro & Way, 1979) which have thermogenic effects. Therefore the present study was undertaken to determine whether the adrenal medulla and the pituitary gland play an essential role in producing the hyperthermia induced by TRH in mice and its potentiation by exogenous noradrenaline or adrenaline. TRH and/or CA were administered to groups of either unoperated (normal), adrenal-demedullated or hypophysectomized mice. Plasma catecholamines were determined in these three groups of mice following TRH administration.

Methods

Male CDI mice (Charles River, France), 6 weeks old and weighing 27–31 g were used. The animals were maintained on a 12 h light-dark cycle (08 h–20 h) in a temperature regulated room (22 ± 1 °C).

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Surgery

Bilateral adrenal demedullation was performed under chloral hydrate anaesthesia (400 mg kg^{-1} i.p.). A dorsal midline skin incision was made and then, bilateral enucleation was performed by lifting each adrenal gland carefully, constricting its vascular supply, slitting the capsule with microscissors, and extruding the body of the gland with small forceps. The viability of the adrenal cortex in these adrenal-demudullated mice was demonstrated by their capacity to survive without having to be maintained on saline. Hypophysectomy was performed by suction, using the transauricular approach, under ether anesthesia following the method of Koyama (1962). All hypophysectomized mice were given a 5% glucose solution in place of water for the first 5 days after surgery. Hypophysectomy was confirmed by microscopic examination of the aspirated tissue just after surgery and by visual inspection of the sella turcica at the end of experiments. In addition, cessation of growth and reduction of adrenal weights were taken as indices of hypophysectomy. Operated mice were allowed to recover for 7 days before being tested.

Measurement of core temperature

The rectal temperature was measured with a thermocouple probe (Bailey Instruments) carefully inserted to a depth of 1.5 cm. Equilibration of the colonic temperature readout occurred within 5–10 s after probe insertion. Readings were taken at 60, 30 min and just before, and every 15 min for the 60 min after each injection. Mice were free to move in their cages except during the temperature measurement. All experiments were performed between 09 h 30 min and 13 h 00 min. The results indicated for 9–13 mice were derived from 2 series of experiments.

Drugs

The drugs were dissolved in 0.9% w/v NaCl solution (saline) and were injected intraperitoneally (i.p.) in a volume of 0.1 ml per 10 g body weight. Each experiment was performed on 3 groups of mice: unoperated (normal), adrenal-demedullated and hypophysectomized groups. Each group included controls (saline), animals treated with TRH 40 mg kg^{-1} (TRH synthesized in our laboratory) or with (−)-noradrenaline D-bitartrate monohydrate (Regis) 1 mg kg^{-1} or (−)-adrenaline (Sigma) 1 mg kg^{-1} and animals treated with TRH and noradrenaline (NA) or adrenaline (Ad). For these last experiments, TRH and NA or Ad were injected simultaneously but separately.

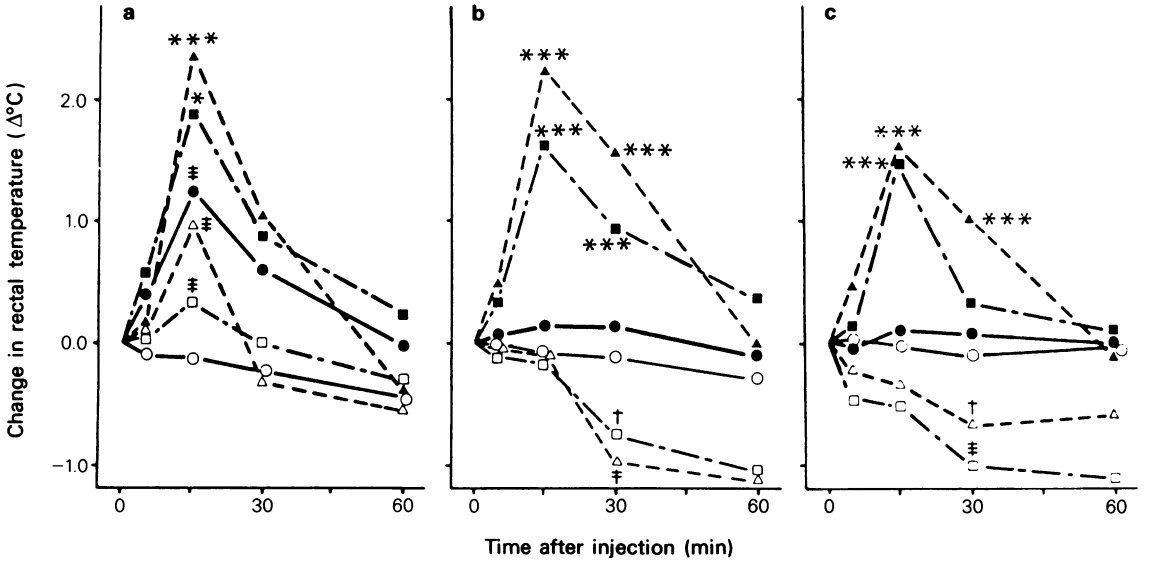

Figure 1 Time course (min) of the rectal temperature change (°C) produced by i.p. injection of (○) saline, (□) noradrenaline (NA, 1 mg kg^{-1}), (△) adrenaline (Ad, 1 mg kg^{-1}), (●) TRH (40 mg kg^{-1}), (■) TRH plus NA and (▲) TRH plus Ad in unoperated (a), adrenal-demedullated (b) and hypophysectomized (c) mice. The thermal responses are expressed as the difference between the temperature just before and the temperatures after injection. Each point represents the mean of 9 to 13 mice. In order to simplify the figures the vertical lines indicating the s.e. mean have been omitted. *$P<0.025$; ***$P<0.001$, compared to TRH-treated mice; †$P<0.025$; ‡$P<0.005$; ‡$P<0.001$; compared to saline-treated mice (Student's t test).

Determination of plasma catecholamines

Approximately 400 μl blood samples were collected from the external jugular vein 15 min after injection. One group was injected with TRH (40 mg kg^{-1} i.p.). Control animals received saline injection. Six to ten mice were used for each determination. Plasma catecholamines were determined in unoperated, adrenal-demedullated and hypophysectomized mice using high performance liquid chromatography (h.p.l.c.) with an electrochemical detector according to the method of Mefford, Ward, Miles, Taylor, Chesney, Keegan & Barchas (1981).

Results

Effects on unoperated mice

Figure 1a shows changes in rectal temperature after TRH injection in unoperated mice. As usual, TRH 40 mg kg^{-1} i.p. produced a rapid significant hyperthermia ($P<0.001$; Student's t test). The peak TRH activity was at 15 min. NA or Ad, both at 1 mg kg^{-1} i.p., administered with TRH (40 mg kg^{-1}) strongly enhanced this hyperthermia 15 min after drug injections. As shown in Figure 2a, in unoperated mice, TRH itself significantly increased plasma NA and Ad in comparison with saline-pretreated mice 15 min after injection.

Effect of adrenal demedullation

To investigate whether the adrenal medulla plays a role in TRH-induced hyperthermia and in its potentiation by exogenous CA, saline, TRH (40 mg kg^{-1}) or Ad or NA (1 mg kg^{-1}) plus TRH were administered to adrenal-demedullated mice. The mice that received TRH alone did not show the usual hyperthermia (Figure 1b). The rectal temperature was not significantly different from that in saline-pretreated mice. However, the simultaneous injection of TRH plus NA or Ad still caused a significant hyperthermia ($P<0.001$) at 15 and 30 min even though NA and Ad alone showed a significant decrease in rectal temperature at 30 min (Figure 1b).

As shown in Figure 2b, plasma Ad in the saline-pretreated mice had almost totally disappeared whereas the plasma NA level was similar to that determined in saline-unoperated mice. In the adrenal-demedullated mice, administration of TRH produced no significant change in the concentration of these plasma catecholamines (Figure 2b).

Effect of hypophysectomy

To investigate whether the pituitary gland is implicated in TRH-induced hyperthermia and in its potentiation, saline, TRH or Ad (or NA) plus TRH were administered to hypophysectomized mice. TRH did not produce the usual hyperthermia (Figure 1c). There was no significant difference in rectal temperature between TRH- and saline-pretreated mice. In contrast, the simultaneous injection of TRH and Ad or NA still produced a significant hyperthermia ($P<0.001$) at 15 min despite the fact that NA and Ad alone showed a tendency to decrease the rectal temperature (Figure 1c). Figure 2c shows that TRH (40 mg kg^{-1}) induced a significant increase in both plasma NA and Ad levels as in the TRH-pretreated unoperated mice.

Discussion

Our previous studies (see Introduction) have demonstrated that, in mice, TRH (40 mg kg^{-1}) produces an increase in body temperature which is directly con-

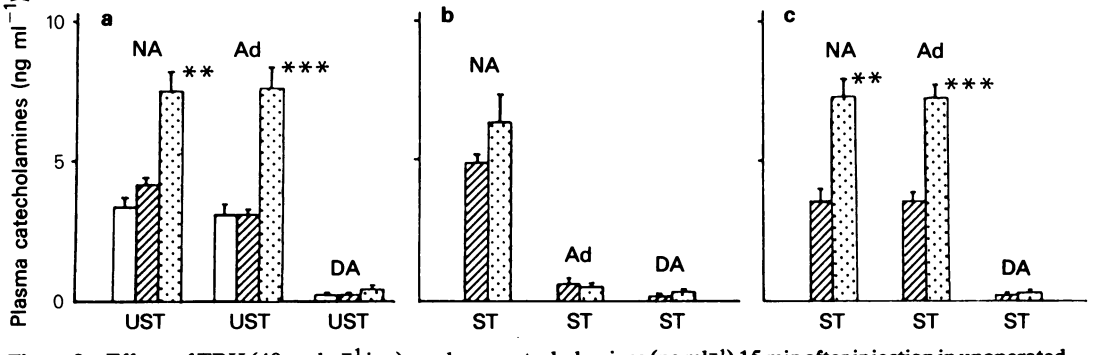

Figure 2 Effects of TRH (40 mg kg^{-1} i.p.) on plasma catecholamines (ng ml^{-1}) 15 min after injection in unoperated (a), adrenal -demedullated (b) and hypophysectomized (c) mice. U: untreated, S: saline, T: TRH, NA: noradrenaline, Ad: adrenaline, DA: dopamine. Each column represents the mean of 6 to 10 samples; vertical lines show s.e. mean. $^{**}P<0.005$; $^{***}P<0.001$; compared to saline-treated mice (Student's t test).

trolled by an α-adrenergic system. The present findings show that TRH induced a rapid short-lasting hyperthermia in normal mice which was accompanied by an increase in plasma noradrenaline and adrenaline levels. In normal mice, noradrenaline or adrenaline administered simultaneously with TRH caused an increase of the TRH-induced hyperthermia.

The main peripheral source of adrenaline is the adrenal medulla whereas the greater part of circulating noradrenaline is released by the sympathetic nerve endings. This is in agreement with our results since adrenal demedullation suppressed plasma adrenaline and did not modify plasma noradrenaline. So, bilateral adrenal demedullation completely abolished the hyperthermia and the increase of plasma catecholamines usually produced by TRH in mice but not the potentiation of this hyperthermia by exogenous catecholamines. This indicates that, to elicit hyperthermia, TRH stimulated the adrenal medulla to increase catecholamine release. Brown (1981) has reported that central injection of TRH in rats resulted in elevation of plasma adrenaline and noradrenaline which may be related to increased heart rate (Tonoue, 1977) and blood pressure (Beale, White & Huang 1977) produced by TRH. It is known that catecholamines have a hyperthermic effect which may be due to the cutaneous vasoconstriction (Jansky, 1979) or to an action on brown adipose tissue (Smith & Horwitz, 1969; Jansky, 1979). In contrast, the potentiation of the TRH-induced hyperthermia was not dependent on the adrenal medulla. This reminds us of the potentiation of TRH-induced hyperthermia by amphetamine (Desiles, Morier & Rips, 1977) which could be of a similar nature. This last potentiation had a different mechanism from that of TRH-induced hyperthermia (Desiles, 1982).

It is quite interesting that TRH in hypophysectomized mice gave the same thermogenic responses as in adrenal-demedullated mice. The hyperthermia elicited by TRH was suppressed in hypophysectomized mice while the mixture of TRH plus noradrenaline or adrenaline induced a considerable hyperthermia. However, unlike adrenalectomy, hypophysectomy did not modify the increase of plasma catecholamines observed following TRH administration. These results indicate that the pituitary gland also plays a role in TRH-induced hyperthermia but not in its potentiation by catecholamines. The fact that in hypophysectomized mice TRH increased the plasma catecholamine levels and did not produce hyperthermia whereas in adrenal-demedullated mice TRH induced neither increase of plasma catecholamines nor hyperthermia suggests that the circulating catecholamines were not solely responsible for the induction of the TRH-induced hyperthermia. A pituitary factor might also be implicated.

In conclusion, the TRH-induced hyperthermia is mediated by a different mechanism from that of the potentiation of this hyperthermia by exogenous catecholamines. The first one is abolished by both adrenalectomy and hypophysectomy, the second is not. The effect of adrenal demedullation on TRH-induced hyperthermia confirms the involvement of an α-adrenergic system but the effect of hypophysectomy does not. An explanation could be that two endogenous substances are necessary to produce hyperthermia induced by TRH in mice: one could be adrenaline (and noradrenaline) released from the adrenal medulla and the second a cofactor secreted by the pituitary gland.

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