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SHORT COMMUNICATION

ADRENAL EPINEPHRINE DEPLETION AFTER INSULIN-INDUCED HYPOGLYCAEMIA IN YOUNG HYPOTHYROID RATS

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We studied the influence of the thyroid hormones on the development of the ability of the adrenal glands to release epinephrine after stimulation by insulin-induced hypoglycaemia in the young rat. In animals rendered hypothyroid from birth, this development is delayed as compared to control rats. Administration of thyroxin to these animals restores the ability of the adrenals to deplete epinephrine. These results may indicate an influence of the thyroid hormones on the development of the functional innervation of the adrenal medulla in the young rat.

Keywords: adrenal gland; epinephrine depletion; insulin hypoglycaemia; neonatal hypothyroidism.

Adrenal epinephrine depletion after insulin-induced hypoglycaemia has long been used in order to study the functional development of the adrenal medulla in the young rat (Slotkin, 1973).

In previous papers we described an acceleration in the development of the adrenal catecholamine content and biosynthesis, due to hypothyroidism in the young rat (Gripois et al., 1980; Gripois and Valens, 1983). In the present study we attempted to determine the influence of neonatal hypothyroidism on the development of the ability of the adrenal glands to secrete epinephrine.

MATERIALS AND METHODS

Young Sherman rats of both sexes were rendered hypothyroid from birth by daily administration of propylthiouracil (PTU; 50 mg/day in 4 ml of water) to their mother by gastric intubation. Control animals received no treatment. A few animals whose mother was given PTU

received daily s.c. injections of thyroxin (T_4; 100, 200 or 250 μg/kg day) (Rastogi and Singhal, 1974).

At specific ages, the litters were divided into two parts: half of the pups received a s.c. injection of saline and half received an injection of insulin (10 IU/kg). After 4 h the rats were killed, and the adrenals dissected and homogenized in 0.4 N perchloric acid. After centrifugation, the supernatant was used for the fluorometric estimation of epinephrine and norepinephrine, according to von Euler and Lishajko (1961). As we confirmed that norepinephrine is not secreted in response to hypoglycaemia, we expressed our results as percentage depletion of epinephrine: for each insulin-treated rat, the percentage depletion was calculated from the mean epinephrine content in the adrenals of the saline-injected pups of the same litter. The statistical significance between the different experimental groups was calculated using the *U*-test of Mann and Whitney.

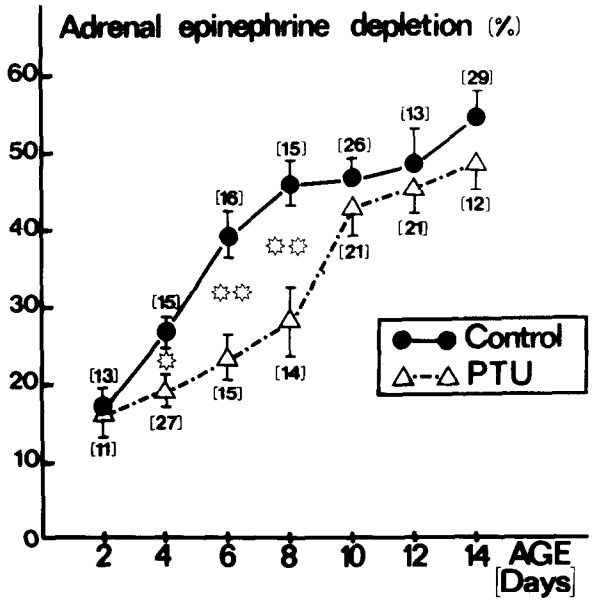

Fig. 1. Intensity of adrenal epinephrine depletion evoked by insulin in developing control and hypothyroid (PTU) rats. Means ± SEM, number of animals in brackets. Significant differences: * $P \leqslant 0.05$; ** $P \leqslant 0.01$.

RESULTS

The evolution of the intensity of adrenal epinephrine depletion in young rats aged 2–14 days is summarized in Fig. 1. In the control animals the percentage depletion of epinephrine increases from 2 to 8 days, then remains constant. In the hypothyroid animals this percentage increases from 2 to 10 days; in these animals it is significantly lower than in the control rats at 4, 6 and 8 days. In addition, the adrenal epinephrine content of the saline-injected pups is given in Table 1.

Treatment with thyroxin (Fig. 2) at a dose of 250 μg/kg/day restores the ability of the adrenals to deplete epinephrine in the PTU-treated pups at the age of 6 days. This indicates no direct effect of PTU, even if, in our experimental conditions, a rather high dose of thyroxin is necessary to restore the adrenal secretion pattern.

DISCUSSION

The administration of PTU to a suckling female rat is well known to produce a clear state of hypothyroidism in the pups (Barrnett, 1950; Schalock et al., 1979). Moreover, in the perinatal period this effect of PTU can be detected 3–4 days after the beginning of the treatment (Jost, 1957; Maillard, 1959). Thus, the present results clearly show that neonatal hypothyroidism induces a delay in the onset of epinephrine secretion by

Table 1

Adrenal epinephrine content in young control and hypothyroid (PTU) rats

	Age (days)						
	2	4	6	8	10	12	14
Controls	0.60 ±0.04	1.01 ±0.06	1.37 ±0.08	2.11 ±0.04	2.79 ±0.17	3.43 ±0.17	3.76 ±0.04
No. of cases	6	12	12	10	19	8	21
PTU	0.73 ±0.04	1.09 ±0.04	1.17 ±0.08	1.94 ±0.22	2.85 ±0.15	3.11 ±0.04	3.58 ±0.28
No. of cases	8	14	9	10	12	14	12
Statistical significance	$P \leqslant 0.05$	NS	NS	NS	NS	NS	NS

Means ± SEM. NS, no statistical significance.

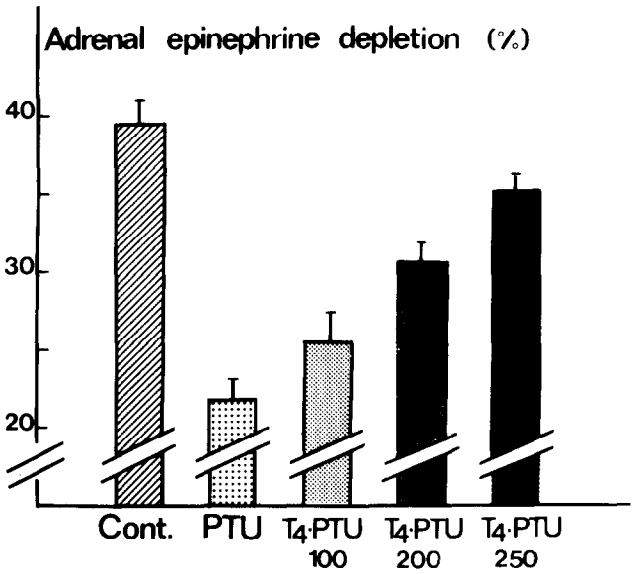

Fig. 2. Influence of thyroxine injections (100, 200 and 250 μg/kg/day) to PTU-treated young rats on adrenal epinephrine depletion at the age of 6 days. Means $\pm$ SEM. Cont., control; PTU, hypothyroid; T_4-PTU, hypothyroid + thyroxine. Statistical significance of the differences: Cont./PTU, $P \leqslant 0.01$; T_4-PTU 100/Cont., $P \leqslant 0.01$; T_4-PTU 100/PTU, NS; T_4-PTU 200/Cont., NS; T_4-PTU 200/PTU, NS; T_4-PTU 250/Cont., NS; T_4-PTU 250/PTU, $P \leqslant 0.01$.

the adrenals in response to a physiological stimulation. In young hyperthyroid rats, opposite effects have been described: thyroxin injection accelerate the development of epinephrine depletion by the adrenals induced by hypoglycaemia (Lau and Slotkin, 1979).

The morphological observations of Mikhail and Mahran (1965) show that an adult pattern of innervation of the rat adrenal medulla is only established by the end of the first week of postnatal life. In the newborn rat, neither insulin hypoglycaemia nor electrical stimulation of the splanchnic nerve can induce epinephrine secretion; on the other hand, direct stimulation of the glands, either in vitro by high K^+ concentrations or in vivo by nicotine, does produce hormonal depletion (Slotkin, 1973; Bartolomé and Slotkin, 1976; Bareis and Slotkin, 1978). These authors concluded that the limiting step in the mechanisms of epinephrine secretion in response to hypoglycaemia in the young rat is the development of the functional connections between the splanchnic nerve and the adrenal medulla (Slotkin, 1973; Bartolomé and Slotkin, 1976, Bareis and

Slotkin, 1978). If they are interpreted in that way, our results show that the thyroid hormones are necessary for the normal development of the functional sympathetic innervation of the adrenal medulla in the young rat.

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