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Hypophysectomy and the sympatho-adrenal system in cold acclimation

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Summary. Hypophysectomy does not impair the increase in weight of brown adipose tissue and adrenals following cold acclimation of the rat. In brown fat, the cold-induced increases in NE and 5 HT contents are not modified by hypophysectomy. In adrenals, hypophysectomy does not change the NE content, but a fall in epinephrine content was observed.
Key words. Cold acclimation; hypophysectomy; brown adipose tissue; adrenals.

Hypophysectomized rats, progressively exposed to a cold environment, are able to produce some heat by non-shivering thermogenesis (NST) and to survive in moderate cold (15 °C)[1]. In spite of the lack of pituitary hormones, cold acclimation also stimulates the development of some characteristics generally induced by cold. Brown adipose tissue (BAT) increases in size and undergoes changes in composition[1] and enzymatic activity[2]. Moreover, hypophysectomy does not prevent the hypertrophy of adrenals which is commonly observed in cold acclimation[3] of the intact rat. BAT is an important site of NST regulated by the sympathetic system and its mediator, norepinephrine (NE)[4] which is found in high concentration in this tissue[5]. It has been reported that serotonin (5 HT), an agent which is also present in high concentration in BAT[5], may also promote thermogenesis[6,7]. It is possible that BAT partly compensates for the absence of pituitary hormones and allows homeothermy at low temperatures. This study was undertaken to determine the levels in BAT and in adrenals of biogenic amines which may have a role in the survival of the hypophysectomized rat in a cold environment.

Materials and methods. Male Long-Evans rats, 6–7 weeks old, were hypophysectomized by the parapharyngeal route. To check the completeness of hypophysectomy, the following criteria were verified: no weight gain, testicular atrophy, examination of the selle turcica in the sphenoid bone under binocular microscopy post mortem. The operation was simulated in another group. After surgery, the rats were maintained at 23 °C for 3 weeks. It was thus possible to check, in vivo, the completeness of hypophysectomy by the absence of resumed body growth. Then both the sham-operated (SO) and hypophysectomized (H) rats were separated into 2 groups, one of which was maintained at 28 °C and the other at 15 °C for 5–6 weeks. Cold acclimation was attained progressively over a 7-day period. The animals were sacrificed by decapitation, adrenals and interscapular brown adipose tissue (IBAT) were excised. All animals were killed between 10.00 and 12.00 h to minimize the effects of circadian changes[8]. Catecholamines and serotonin were extracted from tissues by Bertler's method[9]. Epinephrine (E) and NE contents were determined using the method described by von Euler and Lishajko[10], and 5 HT according to Maickel and Miller[11].

Results and discussion. Hypophysectomy impairs body growth and in hypophysectomized rats, cold acclimation did not modify the body weight. The weight of IBAT decreased after hypophysectomy (by about 40%), but cold acclimation led to an increase of 50% in IBAT weights for both H and SO rats. The amount of IBAT expressed in terms of body weight was greater in the two groups of H rats, 40% for 15 °C, 60% for 28 °C. Adrenal weights were also greatly decreased following hypophysectomy, but were doubled by cold exposure of H animals. These results were in accordance to the previous observations[1]. Thus hypophysectomy does not impair the cold-induced growth of brown adipose tissue and adrenals. The fact that IBAT and adrenals can be increased by gradual cold acclimation of H rat and that these animals are able to produce non-shivering thermogenesis indicates that pituitary dependent hormones are not necessary for the initiation of cold-induced non-shivering thermogenesis. However, deficiency of these hormones may impair the development of the capacity to produce heat at a normal level.

Interscapular brown adipose tissue (table). As previously observed[12,13] the content of NE was higher in cold acclimated rats (50%) than in control rats; hypophysectomy did not influence this cold effect appreciably. Expressed in relative values, there are no significant differences between the four groups.

Effect of cold acclimation on norepinephrine (NE) and serotonin (5HT) contents in interscapular brown adipose tissue (IBAT) and NE and epinephrine (E) contents in adrenals of hypophysectomized rats

	28°C Sham-operated (7)	Hypophysectomized (7)	15°C Sham-operated (7)	Hypophysectomized (5)
a) Interscapular brown adipose tissue				
NE ng/IBAT	167 ± 14	135 ± 9	259 ± 18*	201 ± 15*
NE ng/g	557 ± 24	735 ± 90	548 ± 36	733 ± 36
5 HT ng/IBAT	67 ± 7	54 ± 4	172 ± 23*	151 ± 20*
5 HT ng/g	219 ± 14	293 ± 40	359 ± 36*	519 ± 20*, ×
b) Adrenals				
NE µg/2 adrenals	3.95 ± 0.80	3.32 ± 0.46	3.68 ± 0.98	3.24 ± 1.10
E µg/2 adrenals	28.83 ± 0.96	9.89 ± 0.45×	29.93 ± 1.61	15.24 ± 2.58*, ×

Results are presented as means ± SEM; * significant effect of cold acclimation in SO and H rats; × significant effect of hypophysectomy in rats kept at either 28°C or 15°C; ( ) in brackets: number of animals.

The total and relative values of 5 HT were higher in cold acclimated rats. These results are in close agreement with those previously obtained[14]. Hypophysectomy did not modify the content of BAT in 5 HT; however, the relative level (ng/g) was higher (45%) in H15, than in SO15 rats. It can be concluded that hypophysectomy does not modify the NE and 5HT contents in IBAT and does not impair the cold acclimation-induced increases in these two amine levels.

Adrenals (table). In cold acclimated control rats, although a significant hypertrophy of adrenals was induced, no increases in NE and E contents were observed. Hypophysectomy did not change NE stores in adrenals. However, E content was considerably reduced, 3 times in H28 but only twice in H15; it was significantly more important in this last group than in the former. As indicated by histological studies, the atrophy of adrenals after hypophysectomy is mainly due to atrophy of the zona fasciculata; the medulla is smaller than in normal animals but presents a functional aspect. The fact that hypophysectomy did not affect the NE content but led to a reduction of E content might be explained by a decrease in enzymatic activity which promotes the methylation of NE in E. The activity of phenylethanolamine-N-methyl transferase, an enzyme that synthesizes E from NE in the adrenal medulla, is markedly depressed following hypophysectomy[15,16].

From these results, it may be postulated that the cold-induced stimulation of the sympatho-adrenal system is little modified by hypophysectomy.

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Lack of influence of vitamin D deficiency on insulin release from the isolated pancreatic islets of rats

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Summary. Pancreatic islets were isolated from young (100 g) and adult (390 g), normal and vitamin D deficient male Sprague-Dawley rats. The release of insulin from leucine-stimulated or glucose-stimulated islet was not altered by vitamin D deficiency. The in vitro addition of either 25-hydroxy- or 1,25-dihydroxyvitamin-D had no effect on insulin release from either normal or vitamin D deficient islets. We conclude that the earlier report (Normal et al., Science *209* (1980) 823–825) on vitamin D deficiency depressing insulin secretion from the perfused pancreas must be related to the vitamin's effect on insulin synthesis and not the islet's release of insulin.
Key words. Vitamin D deficiency, insulin release in; vitamin D deficiency, isolated pancreatic islet activity in; Sprague-Dawley rats, vitamin D deficiency in.

Recently Norman et al. reported that vitamin D deficiency depressed insulin secretion from the isolated perfused rat pancreas stimulated by the secretogogues, arginine[1] and glucose[2]. In support, Clark et al.[3] showed that administration of 1,25-dihydroxyvitamin-D to vitamin D deficient rats increased the serum level of insulin.

To test whether vitamin D is required for the synthesis and/or release of insulin from the pancreas, we have focused on the release aspects of vitamin D treatment. 25-Hydroxy- and 1,25-dihydroxyvitamin-D were added to isolated pancreatic islets from young and adult, normal and vitamin D deficient rats, and the amount of insulin released upon stimulation by leucine or glucose measured by radioimmunoassay.

Materials and methods. Individual islets were isolated from the pancreas of young (100 g) and adult (390 g) normal and vitamin D deficient male Sprague-Dawley rats as previously reported[4].