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38

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# Effect of Thyroidectomy on the *in vitro* Utilization of Glucose by Hypophysis and Hypothalamus in the Rat[1]

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*Key Words.* Thyroidectomy · Hypophysis · Hypothalamus · Glucose metabolism

*Abstract.* The *in vitro* utilization of 1-$^{14}$C- and 6-$^{14}$C-glucose was studied in the neuroendocrine structures of the hypophysis and hypothalamus 2 months after thyroidectomy in rats fed a low iodine diet. At this time, their PBI was markedly decreased and TSH levels were increased. Elevated levels of 6-$^{14}$C-glucose uptake and its conversion to $^{14}CO_2$ and $^{14}$C-lactate were obtained in the posterior hypothalamus from the thyroidectomized rats compared to control animals. The formation of $^{14}CO_2$ from 1-$^{14}$C-glucose was increased in both the median and posterior hypothalamus of the thyroidectomized animals. No differences were found between thyroidectomized rats and controls in the uptake of glucose and formation of $^{14}CO_2$ and $^{14}$C-lactate in the hypophysis and in the anterior hypothalamus. The effects of thyroidectomy on glucose metabolism are discussed in terms of the functions of the neuroanatomical structures in the hypothalamus and hypophysis of the rat.

Many close relationships among thyroidal, hypophyseal and hypothalamic functions are well-established [15]. Efforts to establish the metabolic situation of the hypophysis under conditions of altered thyroid status have shown, for example, that thyroidectomy produces an increase in both carbohydrate utilization [12, 14] and protein synthesis [8, 10, 13, 16, 17] in the pituitary and that administration of thryoid hormone suppresses these effects. We became interested in the relationship between the hypothalamus and thyroid status, but were unable to find studies on the metabolism of the hypothalamus in thyroidectomized animals. Therefore, we examined the

[1] Part of this study was carried out at the Departamento de Endocrinologia Experimental, Instituto G. Marañón, CSIC. Madrid.



*in vitro* utilization of 1-$^{14}$C- and 6-$^{14}$C-glucose by different regions of the hypothalamus from thyroidectomized rats.

## *Materials and Methods*

50–60 g Wistar female rats were surgically thyroidectomized. 2 days later, an intraperitoneal injection of 50 $\mu$Ci $^{131}$I-NaI was administered to destroy any remaining thyroid gland. Age- and sex-matched intact animals were used as controls. Thyroidectomized animals were fed *ad libitum* with a low iodine diet containing 0.05–0.09 $\mu$g iodine/g diet [5]. The controls were fed the low iodine diet supplemented with 1.7 $\mu$g $KIO_3$/g diet. All animals drank distilled water *ad libitum.* 50–60 days after thyroidectomy, rats were killed by decapitation. Blood was collected from the neck into heparinized beakers. After centrifugation, PBI [2] and TSH [6] were evaluated in the plasma. Hypophysis and hypothalamus were removed immediately from the head and weighed. Each hypothalamus was dissected into its anterior, median and posterior parts. Tissues were incubated in 0.25 ml of Krebs-Ringer bicarbonate buffer (pH 7.4) containing 1 $\mu$Ci 1-$^{14}$C-glucose or 6-$^{14}$C-glucose (1 mg/ml). After 60 min incubation at 37 °C and shaking (100 cpm), the media was processed for the determination of labelled glucose, $CO_2$ and lactate as previously described [3].

## *Results*

Table I presents the data obtained for the plasma PBI and TSH, and body, pituitary and hypothalamus weights in the intact controls and thyroidectomized rats 2 months after removal of the thyroid gland. Plasma PBI was significantly lower in the thyroidectomized rats than in the controls while plasma TSH was elevated after thyroidectomy. Growth ceased soon after thyroidectomy and at the time of decapitation the thyroidectomized rats weighed significantly less than the controls. The absolute weight of the hypophysis and hypothalamus did not differ between the thyroidectomized and control groups, but when expressed in terms of body weight, the relative weight of the hypophysis and hypothalamus in the thyroidectomized rats was significantly greater than in the controls.

Table II presents the data obtained for glucose metabolism in the hypophysis and hypothalamus in intact and thyroidectomized rats. The uptake of 1-$^{14}$C-glucose and 6-$^{14}$C-glucose and their conversion to $^{14}CO_2$ and $^{14}$C-lactate in the hypophysis and the anterior hypothalamus was essentially the same in thyroidectomized and control animals. In the medium hypothalamus, however, the formation of $^{14}CO_2$ from 1-$^{14}$C-glucose was higher in the thyroidectomized rats than in the controls. The formation of $^{14}CO_2$

*Table I.* Effect of thyroidectomy in the female rat (mean ± SEM)

| | Plasma | | Body weight | Pituitary weight | | Hypothalamus weight | |
|---|---|---|---|---|---|---|---|
| | PBI[1] µg/ml | TSH µg/ml | g | mg | mg/100g body weight | mg | mg/100g body weight |
| Controls | 5.7 ±0.3 (13) | 14 (13) | 183±5 (13) | 10.5±0.4(15) | 5.7±0.2(16) | 46.9±2.3 (16) | 25.5±1.3 (16) |
| Thyroidectomized | 0.28±0.02 (11)* | 1,080 (11) | 99±3 (19)* | 8.4±0.3(14) | 9.3±0.4(14)* | 39.7±2.7 (14) | 44.2±3.4 (14)* |

Number of rats/group in parentheses. * $p < 0.001$ compared to controls. [1] PBI = Protein-bound iodine.

*Table II.* Effect of thyroidectomy on the *in vitro* utilization of (1-$^{14}$C) and (6-$^{14}$C) glucose by whole hypophysis and regions of the hypothalamus from female rats (means ± SEM)[1]

| | Uptake of 1-$^{14}$C-glucose | Formation of $^{14}CO_2$ | Formation of $^{14}$C-lactate | Uptake of 6-$^{14}$C-glucose | Formation of $^{14}CO_2$ | Formation of $^{14}$C-lactate |
|---|---|---|---|---|---|---|
| Hypophysis | | | | | | |
| Controls | 13.3±0.3 (5) | 0.80±0.09 (6) | 6.9±0.6 (6) | 12.6±0.9 (8) | 0.67±0.14 (9) | 7.6±0.7 (7) |
| Thyroidectomized | 12.5±1.0 (8) | 0.79±0.08 (8) | 6.9±0.6 (8) | 12.4±0.7 (7) | 0.52±0.08 (8) | 6.7±0.6 (7) |
| Anterior Hypothalamus | | | | | | |
| Controls | 24.7±2.4 (7) | 0.88±0.12 (8) | 19.4±2.7 (8) | 24.8±2.6 (8) | 0.76±0.20 (8) | 16.9±1.8 (7) |
| Thyroidectomized | 30.1±2.1 (8) | 1.05±0.10 (8) | 20.4±1.1 (7) | 30.5±1.8 (7) | 1.02±0.22 (8) | 21.0±1.4 (8) |
| Median Hypothalamus | | | | | | |
| Controls | 23.4±2.3 (6) | 0.66±0.06 (8) | 17.1±1.7 (8) | 23.6±1.2 (7) | 0.68±0.14 (8) | 16.6±1.2 (6) |
| Thyroidectomized | 25.6±1.8 (8) | 0.98±0.05 (8)* | 18.9±1.5 (8) | 26.6±1.4 (7) | 1.19±0.20 (8) | 18.6±1.5 (8) |
| Posterior Hypothalamus | | | | | | |
| Controls | 24.1±1.3 (7) | 1.14±0.07 (8) | 16.8±1.2 (7) | 23.7±1.9 (8) | 1.05±0.21 (8) | 15.1±1.7 (7) |
| Thyroidectomized | 26.0±1.4 (7) | 1.55±0.16 (8)* | 18.2±1.0 (7) | 30.6±1.9 (7)* | 1.88±0.33 (8)* | 21.1±1.2 (8)* |

Number of rats/group in parentheses. * $p < 0.05$ compared to controls.
[1] The data are expressed as the percentage of initial radioactivity in the incubation medium/10 mg tissue.



from 6-$^{14}$C-glucose also tended to be higher in the medium hypothalamus of thyroidectomized rats than in controls, but the difference was not statistically significant. In the posterior hypothalamus, the formation of $^{14}CO_2$ from 1-$^{14}$C-glucose and the uptake of 6-$^{14}$C-glucose and its conversion to $^{14}CO_2$ and $^{14}$C-lactate were significantly enhanced in the thyroidectomized animals.

### *Discussion*

An intense degree of hypothyroidism was present in our thyroidectomized rats, as shown by their early ceasation of growth, low plasma PBI level and high TSH values. Despite this, glucose utilization in the hypophysis of the thyroidectomized animals failed to differ from the level observed in the intact control animals. Our results are in apparent contradiction with those of Matsuzaki [12] who found an augmented utilization of glucose by the hypophysis of thyroidectomized rats. The different results could be explained by the fact that our study was carried out 2 months after thyroidectomy while Matsuzaki used animals 2 weeks after thyroidectomy. It is known that opposite metabolic changes are present in the hypophysis of thyroidectomized rats such as an increase in the synthesis of TSH and prolactin [8, 18], a reduction in that of growth hormone [8] and degeneration of non-TSH-trophic cells [7]. The possibility exists that these compensatory opposite changes are balanced after prolonged time of thyroidectomy to normalize the previously augmented glucose metabolism.

We have seen here that glucose metabolism in the anterior hypothalamus does not differ between the thyroidectomized rats and the controls. This hypothalamic area includes the preoptic area and the suprachiasmatic nucleus, [4, 15], where few TRH-secreting cells seems to be located [4, 5].

The increased production of $CO_2$ from glucose in the median hypothalamus from the thyroidectomized rats might be related to the role of this region in the secretion of TRH: the median hypothalamus includes the paraventricular nucleus and the nerve fibres that go through the median eminence where most TRH-secreting cells are located [1, 9, 11, 15]. The high TSH levels in the plasma of the thyroidectomized rats suggests that the TRH-secretion in these animals was elevated. Perhaps the augmented metabolic activity in the median hypothalamus of the thyroidectomized rats reflects an enhanced activity of neuroendocrine structures for the production and secretion of TRH.

The augmentation of glucose utilization observed in the posterior hypothalamus from the thyroidectomized animals was unexpected. This area contains the mammillothalamic tract and the interpeduncular nucleus [4, 15] where no TRH-secreting cells nor areas giving positive TSH responses following electrical stimulation are located [9, 15]. Based on our findings, however, the posterior hypothalamus appears to play a role in the neuroendocrine physiology of the thyroidectomized animals. The relative contribution of the cell types in the posterior hypothalamus on glucose utilization in the presence and absence of the thyroid gland remains to be investigated in order to clarify the functions of this region under conditions of altered thyroid status.

## *References*

1 AVERILL, R. L. W.; PURVES, H. D., and SIRETT, N. E.: Relation of the hypothalamus to anterior pituitary thyrotropin secretion. Endocrinology *69:* 735–745 (1961).

2 BENOTTI, J. and BENOTTI, N.: Protein-bound iodine, total iodine and butanol extractable iodine by partial automation. Clin. Chem. *9:* 409–416 (1963).

3 CASTRO, M. and HERRERA, E.: Effect of thyroidectomy on circulating components and liver metabolism in fed and fasted rats. Hormone Res. *4:* 357–366 (1973).

4 DE GROOT, J.: The rat hypothalamus in stereotaxic coordinates. J. comp. Neurol. *113:* 389–400 (1959).

5 ESCOBAR DEL REY, F.; MORREALE DE ESCOBAR, G.; JOLIN, T., and LÓPEZ QUIJADA, C.: Effects of small doses of thyroid hormones on thyroid weight in hypothyroid rats. Endocrinology *83:* 41–50 (1968).

6 GARCÍA, M. D.; CACICEDO, L., and MORREALE DE ESCOBAR, G.: Radioimmunoassay for rat TSH. Abstracts. 4th Meet. eur. Thyroid Ass., Berne 1971.

7 GRIESBACH, W. E. and PURVES, H. D.: The significance of the basophil changes in the pituitary accompanying various forms of thyroxine deficiency. Br. J. exp. Path. *26:* 13–17 (1945).

8 IEIRI, T.: Effects of thyroidectomy and triiodothyronine ($T_3$) on the synthesis and release of growth hormone (GH) and prolactin Jap. J. Physiol. *21:* 551–562 (1971).

9 KRULICH, L.; QUIJADA, M.; HEFCO, E., and SUNDBERG, D. K.: Localization of thyrotropin-releasing factor (TRF) in the hypothalamus of the rat. Endocrinology *95:* 9–17 (1974).

10 LEE, K. L.; BOWERS, C. Y., and NEAL MILLER, O.: Some studies of the effect of thyroid hormone on the metabolism of the anterior pituitary gland of rats. Endocrinology *83:* 754–762 (1968).

11 MARTIN, J. B.; BOSHANS, R., and REICHLIN, S.: Feedback regulation of TSH secretion in rats with hypothalamic lesions. Endocrinology *87:* 1032–1040 (1970).

12 MATSUZAKI, S.: Effects of thyroidectomy and thyroxine supplement on the glucose metabolism in the rat anterior pituitary. Endocrinol. Jap. 16*:* 507–515 (1969).

13 MATSUZAKI, S.: Effects of graded doses of L-thyroxine on $^{14}C$-leucine incorporation into anterior pituitary proteins of thyroidectomized rats. Endocrinol. jap. *17:* 379–385 (1970).



14 MATSUZAKI, S.; SUZUKI, M., and MATSUZAWA, T.: Changes in lactate dehydrogenase isozyme patterns of rat anterior pituitary in varied thyroid states. Endocrinol. jap., *16:* 301–308 (1969).

15 REICHLIN, S.; MARTIN, J. B.; MITNIK, M. A.; BOSHANS, R. L.; GRYMM, Y.; BOLLINGER, J.; GORDON, J., and MALACARA, J.: The hypothalamus in pituitary-thyroid regulation. Recent Prog. Horm. Res. *1971:* 229–286.

16 TONOUE, T. and YAMAMOTO, K.: Effect of thyroidectomy and a thyroxine supplement on amino acid incorporation into proteins of rat anterior pituitary. Endocrinology *81:* 1029–1032 (1967).

17 TONOUE, T. and YAMAMOTO, K.: Amino acid uptake and incorporation into rat pituitary as effected by thyroidectomy and thyroxine administration. Biochem. biophys. res. Commun. *26:* 315–319 (1967).

18 TONOUE, T. and YAMAMOTO, K.: Effect of thyroxine on L-alanine incorporation into proteins of the secretory granules of rat anterior pituitary. Jap. J. Physiol. *18:* 481–488 (1968).

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