HISTOPHYSIOLOGICAL STUDY OF THE EFFECT OF CHRONIC EXPERIMENTAL STRESS AND ENDOGENOUS OPIOIDS ON THE THYROID GLAND

R. A. Krasnoperov, V. A. Glumova, S. N. Ryashchikov, and N. É. Proshutina

UDC 616.441-091+616.441-008.6/02:616.45-001.1/3:616-003.725

KEY WORDS: thyroid gland; stress syndrome; endogenous opioids

The functional state of the endocrine organs during development of the stress response largely depends on interaction between the systems responsible for realization and limitation of that response [9]. Data on the effect of an altered emotional status on the pituitary-thyroid complex and of its component — the thyroid gland (TG) — are quite contradictory [2-4]. The stress-realizing effect of catecholamines is known to be largely blocked by stimulation of the group of endogenous opioids [9, 14]. Questions of interaction between organs responsible for the development of the general adaptation syndrome are very important in connection with interpretation of the mechanisms of development of neuroendocrine disturbances.

The aim of the present investigation was to study the histophysiological parameters of TG during emotional stress and additional involvement of the systems limiting the stress reaction.

EXPERIMENTAL METHOD

Experiments were carried out on 15 noninbred rabbits weighing 2.0-2.5 kg, divided into three equal groups. In the animals of experimental group 1 chronic stress was induced by stimulation of the ventromedial nucleus of the hypothalamus by a subthreshold electric current generated by an ÉSL-2 stimulator for 30 days (15 hourly sessions on alternate days) [8]. Besides this stressor excitation, rabbits of group 2 were subjected also to electrical stimulation of the nucleus magnus raphe (NMR) in the medulla, one of the central points for synthesis of opioid peptides [12, 13]. Coordinates of the stimulated brain structures were found on a stereotaxic atlas [6]. The animals of group 3 served as the control. TG was fixed in 10% neutral formalin and embedded in paraffin wax. Sections 5-6 μm thick were stained with hematoxylin and eosin and subjected to stereometric analysis, with determination of the bulk density of the structural components, the height of the thyrocytes, and the mean diameter of the follicles [1]. To assess the response of the microcirculatory bed the vascularization index of the thyroid epithelium was calculated as the ratio of the density of the vascular component to the volume content of the thyrocytes. The volume of the nuclei of the follicular cells was approximated by the equation for the volume of an ellipse of rotation. Serum levels of the following hormones were tested by radioimmunoassay: tri-iodothyronine (T_3), thyroxine (T_4), and thyrotrophin (TTH) by means of commercial kits from "Amersham International" (Great Britain) and "Mallinckrodt Diagnostica" (Germany). Fragments of TG for electron-microscopic analysis were fixed in 3% glutaraldehyde and 1% osmium tetroxide and embedded in a mixture of the resins Epon and Araldite M. Ultrathin sections were studied on the JEM-100C electron microscope (Japan). All numerical results were subjected to statistical analysis by Student's test of differences of arithmetic means.

Department of Biology, Department of Pathological Anatomy, and Department of Normal Physiology, Izhevsk State Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR A. P. Avtsyn.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 113, No. 1, pp. 33-35, January, 1992. Original article submitted February 13, 1991.

TABLE 1. Comparative Characteristics of Morphofunctional Parameters of TG in Control and Experimental Rabbits ($\bar{X} \pm S_{\bar{x}}$)

Parameter	Group		
	1-	2-	3-
Bulk density of thyrocytes, %	53,0±2,29*	53,0±1,58*	66,3±0,43
Bulk density of colloid, %	9,8±0,9	9,9±1,02	8,7±0,25
Bulk density of stroma, %	35,0±2,48*	36,0±1,9*	24,0±0,38
Bulk density of vessels, %	24,0±2,97*	26,0±1,8*	9,0±0,2
Vascularization index, conventional units	0,45±0,01*	0,5±0,01*	0,14±0,01
Diameter of follicles, μ	37,0±0,84*	31,0±0,64	32,3±0,5
Height of thyrocytes, μ	10,1±0,2*	8,3±0,1*	10,9±0,2
Volume of thyrocyte nuclei, μ^3	83,9±1,9*	26,0±1,45*	103,7±2,8
T_3, nmoles/liter	4,47±0,69*	2,3±0,1*	1,24±0,02
T_4, nmoles/liter	5,91±0,02	6,0±0,13	5,95±0,03
	21,0±1,69*	34,9±3,17*	6,2±0,58

Legend. *$p < 0.001$) Significance of differences compared with control animals.

EXPERIMENTAL RESULTS

Under conditions of chronic experimental stress signs of hyperfunction of TG were found: an increase in the serum T_3 concentration (in group 1 by 3.6 times, and in group 2 by 1.8 times compared with the control), whereas the variations in the T_4 level were not statistically significant (Table 1). Changes in the ratio of the thyroid hormones toward an increase in the profile of T_3, hormonal activity of which is 5-6 times greater than the corresponding value for T_4 [10], can be detected in some forms of diffuse toxic goiter [5]. These observations are in agreement with modern views on the important role of emotional trauma in the etiology of the hyperthyroid state [7].

The morphometric analysis of TG of the experimental animals revealed a decrease in bulk density and height of the epithelial lining of the structural units of the gland (adenomers) and also in the mean volume of the thyrocyte nuclei, evidence of inhibition or exhaustion of the follicular epithelium. Under these circumstances the considerable imbalance of the thyroid status of the stressed rabbits, in the direction of hyperfunction (group 1), and the smaller increase in the T_3 level accompanying additional involvement of the opioid peptide system (group 2), point to qualitative differences in the functional state of TG in the groups. Whereas in experimental group 1 reduction of the mean volume of the nuclei and height of the thyrocytes (Table 1) can be regarded as exhaustion of the physiological powers of the thyrocytes by excessive hormone production, in series 2 the corresponding changes must be assessed as signs of inhibition of thyroid function by endogenous morphinelike substances. The changes revealed in the parenchyma of TG of the experimental rabbits take place against a background of a marked vascular response: the vascularization index of the gland exceeded the corresponding value for the control animals by 3.3 and 3.7 times in animals of groups 1 and 2 respectively (Table 1). Direct correlation between the parameters of the blood supply to TG and the TTH profile points to a vasotropic effect of the latter. At the microscopic level signs of stasis and sludging of erythrocytes were seen (Fig. 1), together with transudation of plasma, an increase in density of the vascular component, edema of the stroma, and dilatation of lymphatics (Fig. 2). Desquamation of the epithelium in the cavity of the follicles and disturbance of the basement membrane of the adenomers were observed in sections through TG in both experimental groups. Changes brought about by the action of the negative emotional factor were more marked in group 1. The reason is evidently activation of the opioid peptide complex in response to stimulation of NMR (group 2), causing limitation of stress-induced damage to the glandular structures.

The histophysiological changes in TG of the experimental animals described above were accompanied by activation of stimulation of the thyroid parenchyma by TTH, whose level was 3-5 times higher than in the control group (Table 1). Elevation of the serum TTH profile in the presence of additional stimulation of the opioid-synthesizing zones of the brain (group 2) was more marked than in group 1. Consequently, the rising level of opioid peptides in the CNS during stimulation of NMR [12, 13] stimulates TTH secretion, in agreement with data in the literature [11]. Nevertheless, TG of the rabbits of group 2, activated by TTH, responds by a smaller degree of internal secretion of T_3 than in group 1, and is characterized by marked morphological signs of inhibition. The effect of the pituitary inducer on TG of the animals of group 2 is evidently partially blocked. Since endogenous opioids, because of their polypeptide nature, cannot pass through the blood-brain barrier [9], it can be postulated that TG contains its own peripheral stress-limiting mechanisms.

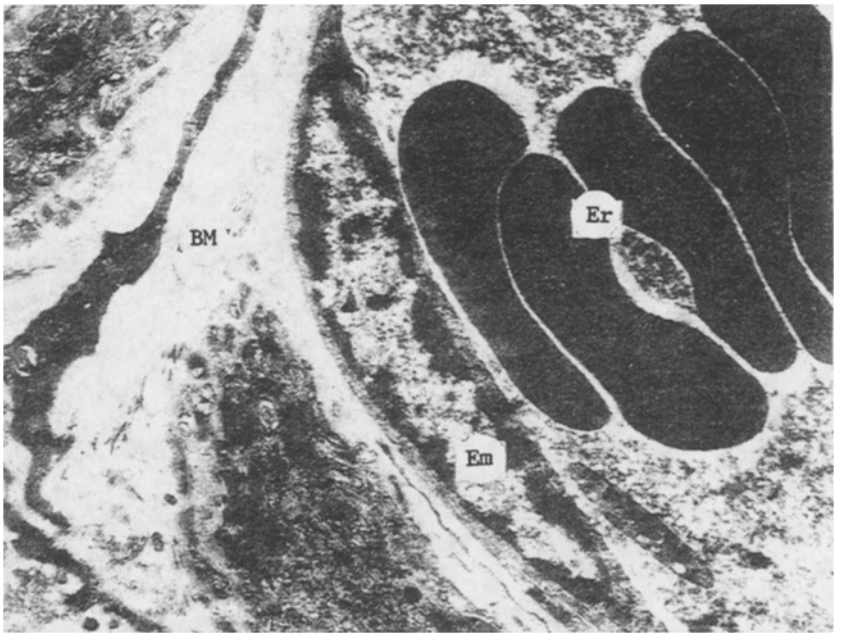

Fig. 1. TG of rabbits of group 1 with edema of basement membrane (BM) and sludging of erythrocytes (Er) in lumen of vessels. Magnification 6000. En) Endothelium.

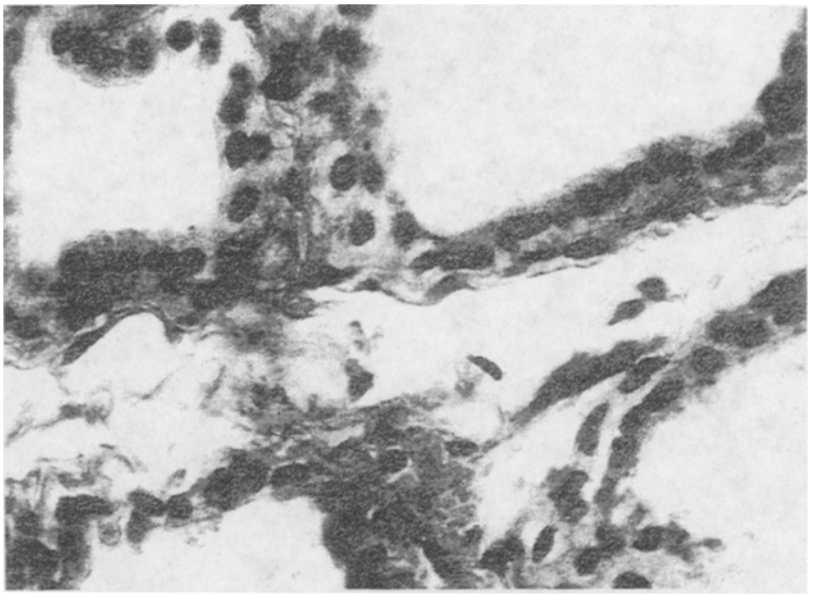

Fig. 2. TG of rabbits of group 1 with signs of deformation of lymphatics, edema and swelling of stroma. Hematoxylin and eosin. Objective 40, ocular 7.

Under conditions of experimental stress, TG thus responds by increased secretion of T_3 into the circulation even though changes in the T_4 level are not statistically significant. The conversion of this process to the chronic form leads to morphological features of exhaustion of TG. Additional activation of the opioid peptide complex of the stressed rabbits in this case induces a more marked increase in the concentration of the specific pituitary stimulator in the blood stream.

LITERATURE CITED

1. G. G. Avtandilov, Medical Morphometry [in Russian], Moscow (1990).
2. Ya. I. Azhipa, Nerves of the Glands of Internal Secretion and Mediators in the Regulation of Endocrine Functions [in Russian], Moscow (1981).

3. M. G. Aliev, A. V. Rzaeva, and O. I. Rybakova, Probl. Éndokrinol., No. 5, 74 (1987).
4. E. S. Argintaeva, V. D. Slepushkin, and G. K. Zoloev, Patol. Fiziol., No. 1, 36 (1988).
5. M. I. Balabolkin, Endocrinology [in Russian], Moscow (1989).
6. J. Bures, M. Petran, and I. Zachar, Electrophysiological Methods of Investigation [Russian translation], Moscow (1962).
7. S. Zoografski, Endocrine Surgery [in Russian], Sofia (1977).
8. D. Ganten, N. A. Nikolov, and K. V. Sudakov, Science and Mankind. Annual Review [in Russian], Moscow (1986), p. 25.
9. M. G. Pshennikova, Patol. Fiziol., No. 3, 85 (1987).
10. Ya. Kh. Turakulov, Probl. Endokrinol., No. 5, 78 (1986).
11. H. Matsuo, Exc. Med. Int. Congr. Ser., **652**, 126 (1984).
12. D. J. Mayer and D. D. Price, Pain, **2**, 379 (1976).
13. D. J. Mayer, D. D. Price, and A. Rafil, Brain Res., **121**, 368 (1977).
14. M. Bellet, J. L. Elghozi, and P. Meyer, New Endocrinology, Amsterdam (1979), p. 365.