LITERATURE CITED

1. E. V. Golanov and L. V. Lakyuzhnyi, Dokl. Akad. Nauk SSSR, 242, 469 (1979).
2. L. V. Kalyuzhnyi and E. V. Golanov, Byull. Éksp. Biol. Med., 88, 240 (1979).
3. K. V. Sudakov, E. A. Yumatov, M. E. Chervyakov, et al., Patol. Fiziol., No. 5, 58 (1977).
4. F. Comelli, Acta Anaesth. Ital., Suppl. 26, 99 (1975).
5. D. Fung, J. Hwang, S. Chan, et al., Exp. Neurol., 47, 367 (1975).
6. J. Marx, Science, 195, 471 (1977).
7. D. Mayer and D. Price, Pain, 2, 379 (1976).
8. D. Mayer, D. Price, and A. Rafii, Brain Res., 121, 368 (1977).
9. B. Pomeranz, R. Cheng, and P. Law, Exp. Neurol., 54, 172 (1977).
10. D. V. Reynolds, Science, 164, 444 (1969).
11. D. L. Rhodes and J. Liebeskind, Brain Res., 143, 231 (1978).
12. E. Fifkova and J. Marsala, Stereotaxic Atlases for the Cat and Rabbit Brain, Prague (1960).

EFFECT OF THYROTROPHIC HORMONE ON THE MEMBRANE POTENTIAL OF THYROID GLAND CELLS AND ON THYROID HORMONE SECRETION DURING AGING

E. N. Gorban' and G. V. Valueva

UDC 612.67:612.441.014.423.018.2:612.433.441:591.147.1

KEY WORDS: aging; endocrine system; thyroid gland; membrane potential; secretion of thyroid hormones.

Much factual evidence has now been obtained on age changes in thyroid function and their role in the mechanism of aging [2, 3, 5]. However, the biophysical properties of the membranes of secretory cells of the thyroid gland, which play an important role in the maintenance of cell metabolism, transmembrane transport of materials, and regulation of protein biosynthesis and the cell energy balance, have been inadequately studied [1, 4, 6-8].

The object of this investigation was to study correlation between changes in the membrane potential (MP) of the follicular epithelial cells of the thyroid gland and secretion of thyroid hormones under the influence of thyrotrophic hormone (TTH) during aging.

EXPERIMENTAL METHOD

Experiments were carried out on isolated thyroid glands of male Wistar rats of two age groups: adult (7-12 months) and old (27-32 months). The value of MP of the follicular epithelial cells, the total thyroxine (T_4) level in the blood, and the concentration of cyclic AMP in the thyroid gland tissue were determined. Isolated thyroid glands were perfused with Krebs–Henseleit solution (t = 37°C; pH 7.3-7.4), aerated with a mixture of 95% O_2 and 5% CO_2. MP was recorded by a standard microelectrode technique. Glass microelectrodes filled with 2.5 M KCl solution, with a resistance of 15-30 MΩ and a characteristic tip potential of not more than −5 mV were used. Total T_4 in the blood was determined by means of "Thyropac-4" radioisotope kits (Radiochemical Centre, Amersham, England). Cyclic AMP in the thyroid gland tissue was determined by means of radioimmunologic kits supplied by the same firm. Thyroid gland function was activated by intravenous injection of TTH in doses of 5 or 0.5 units/100 g body weight. MP of the follicular cells of the thyroid gland and the total T_4 concentration in the blood were investigated 1, 2, and 3 h, and the cyclic AMP concentration in the thyroid gland tissue 10 min, after injection of TTH.

Biophysics Group, Laboratory of Physiology, Institute of Gerontology, Academy of Medical Sciences of the USSR. Laboratory of Radiology and Roentgenology, Institute of Endocrinology and Metabolism, Ministry of Health of the Ukrainian SSR, Kiev.

(Presented by Academician of the Academy of Medical Sciences of the USSR D. F. Chebotarev.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 89, No. 6, pp. 645-648, June, 1980. Original article submitted March 13, 1979.

TABLE 1. Effect of TTH on MP of Follicular Epithelial Cells of Thyroid Gland in Rats of Different Ages at Various Times after Injection

Group of animals	Statistical index	Index MP level of follicular epithelial cells, mV	Dose of TTH, units per 100 g	MP of follicular epithelial cells at various times after injection of TTH, mV		
				1 h	2 h	4 h
Adult (7-12 months)	n_1	8	5	6	7	6
	n_2	214		179	184	170
	$M \pm m$	44,7±0,58		40,3±0,78	38,5±0,74	37,7±0,79
	Δ			—4,4	—6,2	—7,0
	P			<0,001	<0,001	<0,001
	n_1		0 5	6	6	6
	n_2			121	146	124
	$M \pm m$			44,2±0,78	43,9±0,74	44,0±0,56
	Δ			—0,5	—0,8	—0,7
	P			>0,05	>0,05	>0,05
Old (27-32 months)	n_1	9	5	6	6	6
	n_2	261		201	179	187
	$M \pm m$	40,3±0,43		40,4±0,35	39,6±0,58	37,9±0,42
	Δ			+0,1	—0,7	—2,4
	P			>0,05	>0,05	<0,01
	n_1		0 5	6	6	7
	n_2			128	120	164
	$M \pm m$			40,1±0,57	39,9±0,46	39,7±1,97
	Δ			—0,2	—0,4	—0,6
	P			>0,05	>0,05	>0,05

Legend. n_1) Number of animals; n_2) number of cells investigated; Δ) gradient of changes compared with initial level; P) significance of differences from initial value of MP.

TABLE 2. Total T_4 Concentration (in $\mu g\%$) in Blood of Adult and Old Rats at Different Times after Injection of TTH

Group of animals	Statistical index	Total T_4 level in blood of intact animals	Time of investigation, h	Dose of TTH, units/100 g	
				0,5	5
Adult (7-12 months	$M \pm m$	5,7±0,7	1	6,8±0,9	10,8±1,3
	P			>0,05	<0,001
	$M \pm m$		2	7,1±0,9	9,7±0,9
	P			>0,05	<0,001
	$M \pm m$		3	7,45±1,0	9,8±1,1
	P			>0,05	<0,01
Old (27-32 months)	$M \pm m$	4,2±0,2	1	4,2±0,9	3,8±0,7
	P			>0,05	>0,05
	$M \pm m$		2	4,5±0,3	5,0±1,1
	P			>0,05	>0 05
	$M \pm m$		3	6,9±0,6	6,8±1,1
	P			<0,001	<0,02

Legend. P) Significance of differences compared with initial level.

EXPERIMENTAL RESULTS

With age, MP of the follicular epithelial cells of the rat thyroid gland was shown to decrease (Table 1).

During stimulation of the secretory activity of the thyroid gland by injection of TTH in a dose of 5 units/100 g body weight, a fall in the value of MP was observed during the first 3 h after injection of TTH in both adult and old rats: In the adult animals a significant fall in MP was detected as early as 1 h after injection, but in the old rats this did not occur until after 3 h. When TTH was given in one-tenth of the previous dose (0.5 unit/100 g body weight), no significant changes in the MP level of the follicular epithelial cells were found in either the adult or the old animals during the first 3 h after injection.

Parallel with the study of MP of the thyroid follicular epithelial cells, the total T_4 concentration in the blood was determined during the first 3 h after injection of TTH. Distinct differences were found between the total T_4 levels in the blood of animals of different ages in response to injection of TTH in a dose of 0.5 unit/100 g body weight (Table 2).

TABLE 3. Effect of TTH on Cyclic AMP Concentration (in nanomoles/mg) in Thyroid Gland Tissue of Rats of Different Ages

Group of animals	Statistical index	Intact animals	After injection of TTH in a dose of 0.5 units/100 g	After injection of TTH in a dose of 5 units/100 g
Adult (7-12 months)	$M \pm m$	4,3±0,3	5,1±0,9	9,3±1,1
	P_1		>0,05	<0,001
Old (27-32 months)	$M \pm m$	2,7±0,2	3,1±0,5	3,7±1,0
	P_1		>0,05	>0,05
	P_2	<0,05	>0,05	<0,01

Legend. P_1) Significance of differences from the value for intact animals; P_2) significance of differences for values in adult and old animals.

First, in the old animals after injection of TTH in a dose of 0.5 unit/100 g body weight, there was a marked increase in the total T_4 concentration in the blood, but this was not found in the rats aged 7-12 months. Second, injection of TTH into the old animals in a dose of 5 units/100 g caused no further increase in the total blood T_4 level compared with that after TTH in a dose of 0.5 unit/100 g. In the adult animals a marked increase in the blood T_4 level was observed after injection of TTH in a dose of 5 units/100 g. Third, the increase in the T_4 concentration after injection of TTH in either of the two doses used took place in the old animals, unlike in the adult rats, only after 3 h, whereas in the adult group the total blood T_4 level was significantly increased as early as 1 h after injection of TTH in a dose of 5 units/100 g.

Considering that the action of TTH on the thyroid gland is mediated through cyclic AMP, age differences in the cyclic AMP concentration in intact animals and in animals receiving TTH were studied (Table 3).

In the old rats a significant fall in the cyclic AMP concentration in the thyroid tissue was observed. After injection of TTH in a dose of 0.5 unit/100 g body weight, no differences in the cyclic AMP concentration were found, compared with intact animals. After injection of TTH in a dose of 5 units/100 g body weight the cyclic AMP concentration was significantly increased in the adult rats, but remained almost unchanged in the old rats.

The results of these investigations indicate a definite link between the MP level of the thyroid cells and processes of secretion of thyroid hormones.

The occurrence of membrane depolarization in adult animals 1 h, and in old animals, 3 h, after injection of TTH in a dose of 5 units/100 g body weight coincided with the time of elevation of the total T_4 concentration in the blood. The depolarization thus observed may perhaps be determined by increased permeability of the membrane to sodium ions. Evidence in support of this suggestion is given by data [9] indicating that TTH simultaneously increases the penetration of radioactive sodium into the thyroid gland and activates its secretion.

The absence of changes in MP of the follicular epithelial cells in the old animals in response to injection of a small dose of TTH (0.5 unit/100 g), the absence of any reaction of the adenylate cyclase–cyclic AMP system, in the form of an increase in the cyclic AMP concentration in the thyroid tissue, and the significant increase in the total T_4 concentration in the blood observed against this background, all suggest possible age adjustments in the system of TTH regulation of thyroid function, assuming that the increase in sensitivity of the thyroid tissue to TTH during aging may perhaps be connected with intrathyroid autoregulatory mechanisms lying outside the membrane and responsible for an age decrease in the iodide and I^+ level in thyroid tissue [10, 11]. However, the possibility of a disturbance of the process of activation of adenylate cyclase and of an increase in the activity of phosphodiesterase, cannot be ruled out and this will form the subject of the writers' further study.

LITERATURE CITED

1. B. N. Veprintsev, "Correlation of electrical activity of the nerve cell with RNA synthesis in it and the role of the cell membrane in the regulation of intracellular RNA synthesis," Author's Abstract of Doctoral Dissertation, Pushchino (1971).

2. N. V. Verzhikovskaya, "The thyroid gland and age," Author's Abstract of Doctoral Dissertation, Kiev (1971).
3. G. G. Gatsko, The Endocrine System During Aging [in Russian], Minsk (1969).
4. A. N. Razumovich, Bioenergetic Processes and Aging [in Russian], Minsk (1972).
5. N. V. Svechnikova and N. V. Verzhikovskaya, in: Fundamentals of Gerontology [in Russian], Moscow (1969), p. 140.
6. V. V. Frol'kis, Fiziol. Zh. (Ukr.), No. 2, 221 (1970).
7. V. V. Frol'kis, Biofizika, No. 3, 470 (1974).
8. V. V. Frol'kis (V. V. Frolkis), Experientia, 29, 1492 (1973).
9. D. H. Solomon, Endocrinology, 69, 939 (1961).
10. J. Van Sande, G. Grenier, C. Willems, et al., Endocrinology, 96, 781 (1975).
11. J. A. Williams and S. A. Malayan, Endocrinology, 97, 162 (1976).

ACTION OF GAMMA-AMINOBUTYRIC ACID AND ITS ANALOGS ON SMOOTH MUSCLES OF VEINS

N. P. Erofeev, G. V. Kovalev, N. A. Ivanova, R. S. Orlov, and L. V. Shebeko

UDC 615.31:547.466.3].015.44:612.73:612.134

KEY WORDS: gamma-aminobutyric acid (GABA); GABA analogs; contractile activity; smooth muscles of veins.

The role of gamma-aminobutyric acid (GABA) in the metabolism of mammalian nerve tissue has been demonstrated by many investigations [5, 10-12].

GABA is nowadays regarded as the mediator of inhibition, because it satisfies all the criteria required for mediators of the CNS [12].

Besides the existence of central mechanisms of the action of GABA, peripheral effects of this substance were discovered a little later: on the cardiovascular system, respiration, and organs of the gastrointestinal tract [1, 3].

In experiments on the isolated ileum [13, 14], the inhibitory effect of GABA is accepted to be dominant.

There is, however, an important gap in the study of the action of GABA on the smooth muscle of isolated vessels. This is a subject of particular importance because GABA has been shown to have a depressor effect on the systemic blood pressure and on tone of the cerebral vessels [7, 8].

In the investigation described below, the action of GABA and its agonists (phenibut, phenylpyrrolidone, LGPI-29) on the contractile activity of smooth muscle cells of the isolated rat portal vein was studied.

EXPERIMENTAL METHOD

Experiments were carried out on isolated segments of the albino rat portal vein. Contractile activity of a segment of the vessel was recorded on a high-speed automatic writer by means of a mechanical to electrical transducer of the 6MN-1B mechanotron [2]. The original Krebs–Henseleit solution had the following composition (in millimoles/liter distilled water): NaCl 118, KCl 4.7, $MgSO_4$ 1.2, $CaCl_2$ 2.5, KH_2PO_4 1.2, $NaHCO_3$ 25, glucose 11.

Both initial and test solutions were aerated with a gas mixture consisting of 95% O_2 and 5% CO_2. The temperature of the solution during the experiment was 34-35°C. All drugs (GABA and its analogs) were of

Department of Normal Physiology, San.-Gig. Medical Institute, Leningrad. Department of Pharmacology, Volgograd Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR V. V. Zakusov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 89, No. 6, pp. 648-650, June, 1980. Original article submitted November 13, 1979.

0007-4888/80/8906-0721$07.50