that the cause of the reduction in myocardial contractility under the influence of serum from patients with severe suppurative infection is multifactorial in nature, including not only microbial toxins, but also factors not directly related to the agents of suppurative infection.

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EFFECT OF STRESS AND THE ANTIOXIDANT IONOL ON CATECHOLAMINE SYNTHESIS AND THE DOPAMINE CONCENTRATION IN THE HEART AND ADRENALS

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Activation of the antioxidant systems of the body during adaptation to stress [6] or administration of extrinsic synthetic antioxidants is known to limit the elevation of the blood corticosterone level and exhaustion of brain catecholamines during long-term stress [4] and, at the same time, to prevent stress-induced damage to various internal organs – from the heart and stomach on the one hand, to the brain and retina, on the other [5]. The problem of the role played in the protective effect of antioxidants in stress by their influence on catecholamine metabolism still remains unsolved.

The aim of this investigation was to assess the effect of the antioxidant ionol (dibunol) on synthesis of catecholamines and their concentration in the adrenals and heart during emotional stress.

EXPERIMENTAL METHOD

Experiments were carried out on male Wistar rats weighing 200-220 g. Emotional-painful stress was induced by the method in [10] for 6 h. The animals were killed by decapitation 2 h after the end of exposure to stress. The synthetic antioxidant ionol (2,6-di-tert-butyl-4-methylphenol) was injected intraperitoneally in a dose of 60 mg/kg daily during the 3 days before stress. The catecholamine concentrations in the heart and adrenals of the animals

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TABLE 1. Effect of Ionol on Intensity of Neuronal Uptake of ^{3}H-Noradrenalin, Synthesis of ^{3}H-Catecholamines (in cpm × 10^3/g tissue) and on Catecholamine Concentrations in Rat Heart during Stress (M ± m; n = 12)

Parameter studied	Experimental conditions			
	control	stress	ionol	ionol + stress
Neuronal uptake of ^{3}H-noradrenalin	70,7±4,5	48,6±3,5***	74,6±4,1	137,0±11,3***
Synthesis of ^{3}H-noradrenalin	7,4±0,51	5,2±0,58*	9,3±1,4	34,3±0,43***
Concentration of noradrenalin	0,99±0,19	0,56±0,1*	0,9±0,18	0,73±0,06
Synthesis of ^{3}H-dopamine	1,9±0,08	2,17±0,26	6,33±0,94*	23,14±0,32***
Concentration of dopamine	0,098±0,012	0,17±0,02**	0,3±0,43***	0,2±0,03***

Legend. Here and in Table 2: *p < 0.05, **p < 0.01, ***p < 0.001.

were determined fluorometrically [3]. Neuronal uptake of ^{3}H-noradrenalin (Amersham International, England) and ^{3}H-catecholamine synthesis from tritium-labeled tyrosine (Izotop, USSR) were studied in isolated atria and adrenals. Chromatographic separation of ^{3}H-catecholamines was carried out on columns with Dowex-50 ion-exchange resin (Serva, West Germany) in the sodium form. A mixture of ^{3}H-adrenalin and ^{3}H-noradrenalin was eluted from the columns with 5 ml of 1 M HCl, after which ^{3}H-dopamine was eluted with 5 ml of 2 M HCl [2]. Radioactivity was measured on an SL-30 liquid scintillation counter (Intertechnique, France) with an external standard.

EXPERIMENTAL RESULTS

The data given in Tables 1 and 2 characterize the effect of emotional-painful stress on the neuronal uptake, synthesis, and concentrations of catecholamines in the heart and adrenals, and they reveal two principal factors.

The first is that the influence of stress on some parameters of catecholamine turnover in the heart and adrenals exhibits a definite common feature, namely lowering of neuronal uptake, synthesis, and the concentration of noradrenalin, and in the adrenals, by lowering of the adrenalin concentration also. Meanwhile a threefold increase developed in the concentration of dopamine, synthesis of which was increased in the heart but unchanged in the adrenals.

The fact that the increase in uptake and lowering of the concentration of noradrenalin and adrenalin during stress were regularly accompanied in both organs by dopamine accumulation in agreement with modern views on the metabolic chain of catecholamine biosynthesis can be explained only on the grounds that during stress, when the utilization of catecholamines is increased, their formation is not limited by the first link of that chain, namely tyrosine hydroxylase, which is responsible for converting tyrosine into dopa, but it is limited at the dopamine-β-hydroxylase stage, i.e., the stage of conversion of dopamine into noradrenalin. Ultimately the state of affairs observed in the present experiments arises: mobilization of the catecholamine reserves is combined with a marked increase in the dopamine concentration both in the stress-realizing organ (the adrenals) and in the effect organ (the heart). When the possible physiological importance of dopamine accumulation in the response to strong and prolonged stress is evaluated, it must be recalled that dopamine, an activator of motor activity [18] and a vasodilator and natriuretic agent [19], participates in realization of the adaptive effect of stress and, at the same time, has a very important regulatory influence on the course of the stress-reaction itself. For instance, dopamine inhibits ACTH

TABLE 2. Effect of Ionol on Intensity of Neuronal Uptake of ^{3}H-Noradrenalin, Synthesis of ^{3}H-Catecholamines (in cpm × 10^3/g tissue) and on Catecholamine Concentrations (in μg/g) in Rat Adrenals during Stress (M ± m; n = 12)

Parameter studied	Experimental conditions			
	control	stess	cionol	ionol + stress
Neuronal uptake of ^{3}H-noradrenalin	53,9±2,7	41,2±3,87**	56,3±4,3	82,2±3**
Synthesis of ^{3}H-noradrenalin	7,16±0,6	5,3±0,23**	7,19±0,97	21,3±1,6***
Concentration of adrenalin	402,4±39,0	209±23***	409,6±52,6	326±26
Concentration of noradrenalin	213±26	149±15*	195±19,3	185±24
Synthesis of ^{3}H-dopamine	4,25±0,32	4,3±0,54	4,3±1,1	17,8±1,8***
Concentration of dopamine	2,51±0,29	7,1±0,79***	2,1±0,38	4,93±0,74*

secretion by the pituitary gland [8] and depresses ACTH-induced corticosterone biosynthesis in the adrenals [15]. It thus behaves as a factor limiting the pituitary-adrenal stage of the stress reaction. It has recently been shown that dopamine limits adrenergic activity at the central level [12] and that in rats, which are more resistant to stress, the density of dopamine receptors in the brain is increased [1]. Finally, it has been shown that activation of dopamine receptors by dopamine agonists prevents ulcer formation in the gastric mucosa during stress [17].

The accumulation of dopamine during intensive stress is thus an example of a situation in which the actual realization of the stress reaction leads to activation of factors limiting this reaction and its adverse consequences.

The second factor is that under conditions of physiological rest the antioxidant ionol has not appreciable effect on the parameters of catecholamine biosynthesis in the adrenals that were studied. In the heart it likewise does not affect synthesis or the concentration of noradrenalin, but at the same time, it increases synthesis and raises the concentration of dopamine. Against the background of ionol administration, exposure to stress affects the concentration and biosynthesis of catecholamines differently in many respects from in the control. In fact, under the influence of stress, neuronal uptake and biosynthesis of noradrenalin are not inhibited but, on the contrary, enhanced. The catecholamine concentrations do not fall significantly, and under these circumstances the principal shift observed in this experiment develops, namely an increase in biosynthesis of dopamine by many times and a considerable increase in its concentration in both organs. In the adrenals, for instance, dopamine biosynthesis is increased fourfold and its concentration doubled. In the heart, biosynthesis of this important regulatory amine is increased 11-fold, and its concentration doubled.

These data are essentially evidence that ionol does not change the relations between activities of the principal enzymes of catecholamine biosynthesis, revealed during stress, and consequently, dopamine accumulation is maintained under these conditions. Meanwhile this antioxidant sharply increases the potential capacity of the whole chain of catecholamine biosynthesis, and this enables their normal concentrations to be maintained during stress.

When the probable mechanism of this unexpected effect is assessed, two possibilities must be considered. First, the possibility cannot be ruled out that ionol, acting as an antioxidant, binds free radicals generated at certain stages of catecholamine biosynthesis [9, 11], and thus ensures higher activity of the enzyme chain responsible for this process. Second, we know that ionol penetrates into cell nuclei and, acting at the genetic level, causes an increase in the de novo synthesis of several enzymes, such as glutathione-S-transferase [7, 16]. Since the synthesis of tyrosine hydroxylase, dopamine-β-hydroxylase, and phenylethanolamine-N-methyltransferase, i.e., the principal enzymes involved in catecholamine synthesis, is determined by identical coding sequences of the genome [13, 14], it can be tentatively suggested that ionol activates the expression of these sequences. The increase in population of the enzymes of catecholamine biosynthesis arising as a result of this situation ought to lead to an increase in the potential capacity of this metabolic chain, as was indeed found in the present experiments during stress, when catecholamine utilization rises. Both these hypotheses require experimental verification. Meanwhile the data given above are unequivocal evidence that the known protective effect of the antioxidant ionol and its ability to prevent exhaustion of catecholamine reserves, and the onset of gastric ulcers during stress, lesions arising in other internal organs are not simply the result of inhibition of lipid peroxidation, but are largely determined by activation of catecholamine biosynthesis and the associated increase in accumulation of dopamine, which may play a role in limiting the stress reaction and the severity of stress-induced injuries.

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RESPONSES OF THE ADRENAL CORTEX AND THYROID GLAND OF HYPOPHYSECTOMIZED RATS TO COOLING

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Animals deprived of their pituitary gland continue to survive for a long time although the state of their endocrine glands does not completely recover [2, 4]. Polenov [3], as the result of an analysis of the formation of the Gomori-positive hypothalamo-hypophyseal neurosecretory system during vertebrate phylogeny and ontogeny, postulated the regulatory influence of nonapeptide neurohormones (vasopressin – VP, oxytocin) directly on peripheral endocrine glands without the participation of adenohypophyseal hormones, i.e., by the para-adenohypophyseal route. This method of regulation assumes its greatest importance in stress situations, and in states approaching the pathological. Evidence of the possible direct effect of VP on the adrenal cortex has recently been published [6, 13, 16]. However, the only evidence of this kind of regulation of the thyroid gland has been obtained by research undertaken more than 20 years ago [9, 11]. The study of rats in the early period (during the first week) after total hypophysectomy (HE) made it possible to examine the response of an animal deprived both of the posterior lobe of its pituitary gland (PLP) and of its adenohypophysis. Regeneration of PLP takes place 4 weeks or more after HE, with the formation of a miniature neurohemal organ [14], so that the responses of an animal deprived of its adenohypophysis alone can be investigated.

The aim of the present investigation was to obtain further data on the importance of adenohypophyseal hormones and nonapeptide neurohormones of PLP in the response of the adrenal cortex and thyroid gland of hypophysectomized rats to stress in the form of cooling.

EXPERIMENTAL METHOD

Experiments were carried out on 86 male Wistar rats weighing 130-150 g. The pituitary gland was removed by the trans-sphenoidal approach under ether anesthesia. Completeness of removal of the gland was verified by examination of the sella turcica region after decapitation of the rats. The animals were divided into the following groups: 1) intact (control), 2) 7 days after HE, 3) 7 days after mock HE, 4) 4-7 weeks after HE, 5) 4-7 weeks after mock HE. The mock HE operation involved all stages of HE except extirpation of the pituitary. The animals of all the above groups were used for comparison with animals of the same groups

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