CHANGES IN ASCORBIC ACID CONTENT IN ADRENALS OF RATS WITH EXPERIMENTAL SLEEP DEPRIVATION

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On the 4th day of experimental sleep deprivation the total ascorbic acid content in the adrenals of rats fell by 36% (mainly on account of the reduced form) while its content in whole blood rose by 75%, mainly on account of dehydroascorbic acid.

The decrease in ascorbic acid content in experimental sleep deprivation was observed in both the adrenal cortex and medulla.

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The adrenals play an important role in the mechanism of the stress reaction of the body [2, 6].

The object of this investigation was to study the biochemical characteristics of adrenal tissues in a state of stress produced by prolonged experimental sleep deprivation.

EXPERIMENTAL METHOD

Experiments were carried out on 47 male albino rats weighing 170-300 g. A special apparatus was devised in the department for the sleep deprivation experiments [4]. Rats kept in this constantly revolving apparatus were unable to sleep for several days. During the experiment they were fed on concentrated food placed directly on the floor of the cylinder, while the vessels of water were fixed to its axis. The animals were first habituated to the experimental conditions. Exhaustion of the nervous system developed in the animals on the 3rd-5th day. The rats died from sleep deprivation on the 4th-5th day.

After four days of sleep deprivation the rats were decapitated, and their adrenals were quickly removed, freed from the capsule, and weighed. Control animals were treated in the same way. The ascorbic acid content in the adrenals and blood was investigated colorimetrically by O. D. Kushmanova's microexpress method [1]. The following indices were determined: total ascorbic acid (i.e., the sum of the reduced and oxidized forms) and its reduced form. The difference between the total and reduced ascorbic acid gave the content of dehydroascorbic acid. Ascorbic acid was demonstrated histochemically by Backhus's method [5]. The moisture content of the adrenals was determined both in the control rats and in the rats with experimental sleep deprivation.

EXPERIMENTAL RESULTS

As Table 1 shows, the total ascorbic acid content in the adrenals of the rats after four days of experimental sleep deprivation was lowered by 36% on account of its reduced form. This is consistent with the view that ascorbic acid, when participating in oxido-reductive reactions, is converted into dehydroascorbic acid, the excess of which enters the blood stream from the adrenals. These processes are intensified during adaptive reactions [3].

Correspondingly, in the whole blood of the rats with experimental sleep deprivation the content of total ascorbic acid rose by 74% (mainly on account of dehydroascorbic acid). The decrease in ascorbic acid content in the adrenals on the 4th day of sleep deprivation may evidently be attributed to increased synthesis of mineralocorticoids.

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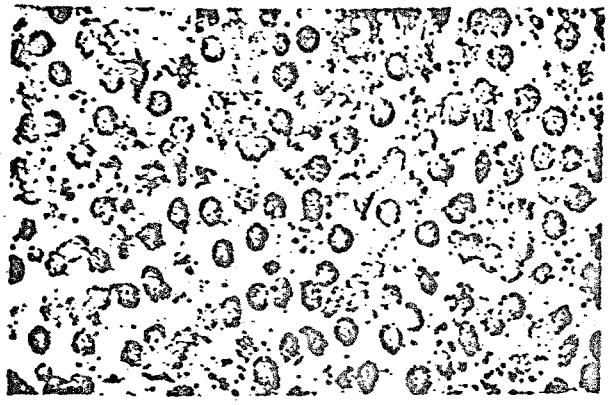

Fig. 1. Distribution of ascorbic acid in adrenal cortex of control rat. Photomicrograph. Objective 8, ocular 10. Backhus's method.

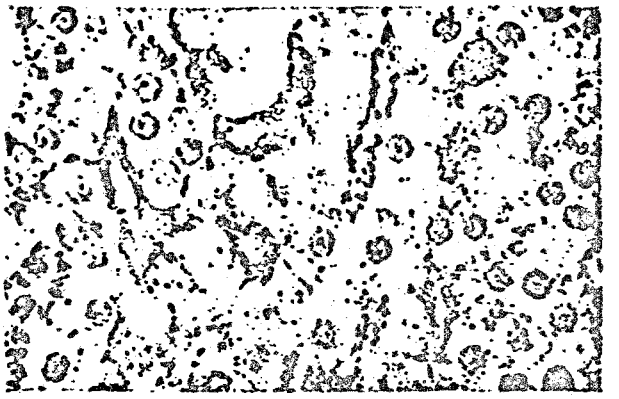

Fig. 2. Liberation of ascorbic acid from glandular cells of adrenal cortex into blood stream in experimental sleep deprivation (4 days). Photomicrograph. Objective 8, ocular 10. Backhus's method.

TABLE 1. Content of Ascorbic Acid in Adrenals and Whole Blood (in μg/100 mg fresh tissue and /100 ml whole blood) in Rats Deprived of Sleep for Four Days

Ascorbic acid	Adrenals					Whole blood				
	control		experimental sleep deprivation			control		experimental sleep deprivation		
	n	M±m	n	M±m	P	n	M±m	n	M±m	P
Total	21	289±9,55	26	180±15,1	<0,001	16	2 205±102	13	4 [illegible]9±334	<0,001
Reduced form	14	171±7,73	25	51±7,6	<0,001	20	1 303±115	29	1 43[illegible]±134	>0,05
Oxidized form	14	181±11,7	21	129±10,3	<0,01	13	679±54,5	15	2 275±[illegible]	<0,001

After four days of experimental sleep deprivation the water content in the adrenals increased on the average by 25.5% (from 67.03±1.47 to 84.2±0.87%; P< 0.001).

In connection with the increased water content in the adrenals of the rats with experimental sleep deprivation, the content of the reduced form of ascorbic acid was expressed per dry weight (31.37 ± 2.06 in the controls, 5.7 ±9.95 μg/100 g dry weight in experimental sleep deprivation; P< 0.001). Under these circumstances the same pattern was observed in the adrenals during sleep deprivation: a decrease in the content of the reduced form of ascorbic acid.

Histochemical methods of determination of ascorbic acid revealed only the reduced form of ascorbic acid in the histological preparations. A high content of ascorbic acid was observed in the control adrenals in all three zones of the cortex. In the zona glomerulosa ascorbic acid was distributed as large granules mainly in the capillary walls and also in the cytoplasm of individual glandular cells. The ascorbic acid content in the zona fasciculata was higher than in the zona glomerulosa. The distribution of ascorbic acid in the zona fasciculata was irregular. However, most glandular cells contained large quantities of it (Fig. 1). In the zona reticularis the distribution of ascorbic acid was more uniform, as numerous tiny granules, present in nearly all the glandular cells. It was found in the adrenal medulla only in solitary chromaffin cells and in the endothelium of the venous sinuses. In all experiments with prolonged sleep deprivation, a redistribution of ascorbic acid took place in the adrenals of the rats, indicating its liberation from the glandular cells into the adrenal blood vessels. In some cells the ascorbic acid granules were larger and were localized at the border between the glandular cells and the walls of adjacent capillaries. In most cells it was absent, and could be seen only in the lumen of the capillaries (Fig. 2). These changes were most marked in the zona fasciculata and zona reticularis of the cortex.

Comparison of the biochemical and histochemical findings thus showed that the reduced form of ascorbic acid in the glandular cells of the adrenal cortex is not only oxidized into dehydroascorbic acid, but also liberated into the blood stream.

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