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SEASONAL RHYTHMS OF INDOLEAMINE LEVELS IN THE RAT PINEAL GLAND

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The role of the pineal gland in the manifestation of biological rhythms is generally familiar. It receives information mainly from the outside world and is an intermediary in interaction between organism and environment, thus enabling the living organism to orient itself to the alternation of day and night and to adapt itself in time. We know that the intensity of serotonin (5-HT) metabolism in pineal cells and formation of its hydroxylated and methoxylated derivatives depend on the intensity of illumination. Data in the literature are evidence that during the dark period of the year (just as during the dark time of the 24-h period) melatonin (MT) production in the pineal gland increases, whereas during the light period of the year (24-h period) it decreases [6, 8, 10] 10]. The opposite data were obtained for 5-HT [4, 12, 14].

Meanwhile the question of the content of other indoleamines in the pineal gland depending on the season has not been adequately discussed in the literature, and it was accordingly decided to study metabolic pathways of 5-HT and concentrations of individual indoleamine fractions in the pineal gland in summer and winter.

EXPERIMENTAL METHOD

Experiments were carried out on 48 mature male Wistar rats weighing 150-200 g. The rats were kept in the animal house under natural conditions of alternation of day and night. The experiments were undertaken at different times of year, with different conditions of daylight: in winter (8 h of daylight and 16 h of darkness) and in summer (16 h of daylight and 8 h of darkness). Levels of the following substances were determined in the pineal gland [12]: 5-HT, N-acetylserotonin (N-AS), MT, 5-methoxytryptamine (5-MOT), and total fractions of 5-hydroxyindoleacetic and 5-methoxyindoleacetic acids (5-HIAA and 5-metOIAA). The animals were decapitated at night (between 1 and 3 a.m., at the time of greatest activity of the pineal gland) in red light. Pineal glands from two or three animals were pooled for indoleamine determination. Fluorescence of the test substances was measured on a BIAN fluorometer at wavelengths of 365 and 470 nm. Weight parameters of the pineal were determined at the same time.

EXPERIMENTAL RESULTS

The results showed that the weight of the pineal gland is not constant but varies with the season of the year. In winter, for instance, the weight of the pineal glands of the rats was 1.61 ± 0.12 mg, whereas in summer it was 1.1 ± 0.08 mg ($P < 0.01$). Parameters of relative weight of the pineal glands showed similar changes.

The results of the biochemical investigation showed (Fig. 1) that the concentrations of 5-HT, N-AS, and MT in the pineal glands of the rats in winter were virtually identical. In summer, however, when the longest period of daylight is observed, the N-AS and MT levels fell abruptly and parallel with one another ($P < 0.001$), whereas the 5-HT concentration in the pineal gland was increased at this time of the year ($P < 0.01$). A different picture was observed when the concentration of 5-MOT was studied, for it increased parallel with the

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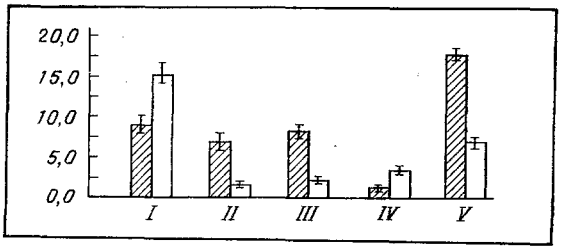

Fig. 1. Concentration of indoleamines in pineal gland (in ng/mg tissue) of mature male rats in summer and winter. I) 5-HT, II) N-AS, III) MT, IV) 5-MOT, V) 5-HIAA + 5-metOIAA. Shaded columns represent winter, unshaded– summer.

lengthening of daylight ($P < 0.001$). As will be clear from Fig. 1, the fraction containing 5-HIAA, 5-metOIAA also underwent sharp fluctuations depending on the season of the year. The concentrations of these substances in the pineal gland was high in winter and sharply reduced in summer ($P < 0.001$).

Assessment of the character of the biochemical changes as a whole indicates that the pineal gland, under physiological conditions, reacts to alternation of daylight and darkness by a change in production and metabolic conversions of all fractions of indoleamines studied. However, these changes differ in direction and are evidence of switching of the pathways of 5-HT metabolism in the pineal gland depending on the changing seasons of the year. In particular, the results suggest that in winter 5-HT is actively metabolized in the pineal gland both via the pathway of N-acetylation followed by orthomethylation with the formation of N-AS and MT, and also via the pathway of oxidative deamination, followed by orthomethylation, with the formation of 5-HIAA and 5-metOIAA, whereas in summer these processes are inhibited. The fact that the 5-MOT level in the pineal gland is minimal in winter and rises sharply in summer suggests that seasonal rhythms of the indoleamine concentration in the pineal gland are determined mainly by the rhythm of N-acetyltransferase activity, for processes of orthomethylation proceed rapidly both in the dark and in the light time of the year. These data confirm the view that the limiting factor of MT formation in the pineal gland during the period of long daylight is in fact N-acetyltransferase [11].

The study of reciprocal relations between metabolic pathways of 5-HT in the pineal gland, via oxidative deamination and N-acetylation appears extremely interesting, for during the study of circadian rhythms in rats it was shown [11] that the first pathway predominates during daylight, with the result that 5-HIAA, 5-hydroxytryptophan, and their methylation products – 5-metOIAA and 5-methoxytryptophan, accumulate in the pineal gland, whereas at night their concentration in the pineal gland falls sharply, but under these circumstances concentrations of N-AS and MT rise. Comparison of data characterizing circadian and seasonal rhythms of indoleamine formation in the pineal gland shows that the process of conversion of 5-HT into N-AS, and subsequently into MT, is controlled by the intensity of illumination, whereas other mechanisms, whose role has received far less study, are involved in the regulation of formation of other 5-HT derivatives in pineal cells.

Data indicating that alternation of the seasons of the year affects changes not only in the intensity of biochemical processes in the pineal cells, but also in the weight of the gland, are of undoubted interest. These data agree on the whole with results obtained by other workers [13] who, in experiments on rats, found an increase in the protein concentration in the pineal gland at night and a decrease by day, due to activation of the β-adrenergic system of the pineal gland in the absence of light. It is considered [5] that in darkness the noradrenalin concentration is increased, as also is the sensitivity of adrenoreceptors, and for that reason the activating system of cAMP, which has a stimulating effect on protein biosynthesis both directly and indirectly, through its action on the processes of its own phosphorylation. This mechanism of an increase in weight of the pineal gland in the dark period of the year seems to the writer to be the most probable, although the effect of MT itself on the intensity of intracellular processes in the pineal cells cannot be ruled out [9].

Much still remains unclear in the interpretation of the physiological purpose of seasonal changes taking places in the pineal gland. At the same time, we know that different hydroxy- and methoxyindoles of the pineal gland differ in their effect on the various stages of function of biological systems in vivo. In particular, MT has a psychotropic action of modulating the cortical inhibitory apparatus and inhibiting the CNS, and it blocks the liberation of hypothalamic releasing hormones and pituitary trophic hormones [3], N-AS selectively inhibits secretion of pituitary thyrotrophin, and thus acts on thyroid function and lowering the concentration of

thyroid hormones accordingly [15], whereas 5-MOT stimulates the resistance of the body by activating the immune system, and it enhances the antitumor effect of irradiation [1], for which reason it has been successfully used, in the form of "mexamine," in clinical oncology.

These sparse and scattered data in the literature are on the whole in full agreement with our own observations, for activation of function of the endocrine glands and potentiation of the protective forces of the body in summer, and their weakening in winter, are well known.

The results of this investigation into changes in metabolic pathways of 5-HT depending on the season of the year provide an approach to the understanding of the mechanism of participation of the pineal gland in the control of biological rhythms.

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