INVESTIGATIVE BEHAVIOR OF RATS DIFFERING IN PREDISPOSITION TO STRESS IN OPEN FIELD AND CHAMBER TESTS

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Distinguishing animals by inborn sensitivity to stress stimuli has served as the basis for their division into stress-resistant (R) and stress-nonresistant (NR) [5, 8]. An important role in the regulation of responses to stress and of inborn forms of behavior is played by the monoamines of the brain [3, 4, 7]. Animals with individual reactivity to stress differ in their relative content and activity of brain monoamines [5, 13]. The problem of how the initial difference in sensitivity to stress and differences in the relative content of biogenic monoamines are reflected in the regulation of certain inborn forms of behavior remains unsolved.

The aim of this investigation was to compare the orienting-investigative behavior and relative levels of activity of the catecholaminergic and serotoninergic systems of the brain in animals depending on the character of their resistance to stress.

EXPERIMENTAL METHOD

Experiments were carried out on 58 male Wistar rats weighing 250-300 g. The animals were first tested for resistance to stress stimulation. A detailed account of the method of testing was given in a previous publication [6]. Investigative behavior was determined in 2 situations: open field testing and burrow chamber testing. The open field, with an area of 1 m^2, divided into 100 small squares, could be used to assess the behavioral features of the animals under conditions of moderate stress, caused by an open space and bright light (a 200-W lamp suspended at a height of 1 m above the center of the field). The animals were under observation during the morning (9-11 a.m.) for 3 min. The number of squares crossed (horizontal investigative activity) and the number of rearings (vertical investigative activity) were counted. The burrow chamber, with a total area of 40 × 40 cm, divided into 16 squares with the same number of holes, could be used to observe behavior of the animals under conditions close to those of their daily life. The number of squares crossed, of rearings, and the number of inspections of the holes were counted. All parameters in the open field and burrow chamber were observed during each minute and during the whole period of observation. At the end of the experiments quantitative biochemical analysis of monoamines and their metabolites was carried out in three brain structures: the frontal region of the neocortex, the hypothalamus, and the caudal part of the brain stem. Concentrations of noradrenalin (NA), dopamine (DA), and serotonin (5-HT) were determined by a modified method in [14]; 5-hydroxyindoleacetic acid (5-HIAA) was determined by the method in [11]; homovanillic acid (HVA) by the method in [15]. The numerical results were subjected to statistical analysis by Student's and the Wilcoxon–Mann–Whitney tests on an "Iskra-226" minicomputer.

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TABLE 1. Concentration of Monoamines and Their Metabolites (ng/g) in Brain Structures of Rats Differing in Resistance to Acoustic Stimulation, %

Group of animals	Brain structure	NA	DA	5-HT	5-HIAA	HVA
Resistant (n = 7)	Cortex	208±30	218±42	318±33	465±89	31±8
	Hypothalamus	228±58	327±58	394±42	613±46	81±33
	Brain stem	290±27	399±49	378±59	808±50	128±26
Nonresistant (n = 7)	Cortex	166±28	413±64*	439±63*	576±149	127±71*
	Hypothalamus	187±18	419±56	317±50	888±62	275±98
	Brain stem	263±25	569±138*	653±57**	918±60	166±24

Legend. Significance of differences between data for rats resistant and not resistant to stress; *$p < 0.05$, **$p < 0.01$ by Student's test; $^{+}p < 0.05$ by Wilcoxon–Mann–Whitney test.

EXPERIMENTAL RESULTS

Comparative analysis of the investigative behavior of the two groups of rats revealed differences in their responses to the open field. Animals resistant (R) to stress differed from the nonresistant (NR) in their higher levels of both horizontal and vertical investigative activity, as shown by an increase in the number of squares crossed and in the number of rearings. The R rats, unlike NR, showed a significant increase in the number of rearings both per minute and for the whole period of observation.

Unlike the open field tests, the behavioral tests in the burrow chamber showed that NR animals had a higher level of horizontal and vertical investigative activity than the R rats, as shown by an increase in the number of squares crossed, the number of rearings, and the number of inspections of holes, both per minute and for the whole period of testing. Thus NR rats in the burrow chamber, unlike in the open field test, exhibited a high level of orienting-investigative behavior. Biochemical analysis of levels of catecholamines, serotonin, and their metabolites in the neocortex, hypothalamus, and caudal part of the brain stem revealed significant differences in their distribution in R and NR rats (Table 1).

The main difference in their distribution was a tendency for the NA level to fall in all structures studied in the NR animals compared with R; this tendency correlated with the raised 5-HT level in the cortex and brain stem, but not the hypothalamus, where the 5-HT level was lowered a little, although the 5-HIAA concentration was increased, indicating intensified 5-HT metabolism in this part of the brain. The DA level was raised in all structures in the NR rats compared with R. A higher DA concentration was observed as the cortical level and also in the brain stem. Meanwhile, in the NR rats the content of the DA metabolite in the brain stem, where the main concentrations of monoaminergic neurons are found, did not differ from the R rats. These findings are in agreement with the results of investigations [5] that showed a deficiency of NA in the brain structures of NR animals compared with R, accompanied by a raised DA level in the brain stem and an increased intensity of 5-HT metabolism in the hypothalamus. It can be tentatively suggested that the decrease in NA, accompanied by an increase in DA, can be explained by reduction of activity of the enzyme dopamine-β-hydroxylase, which catalyzes the transition from DA into NA, in NR rats [5]. Thus the differences observed in the investigative behavior of the NR rats are due to genetically weakened activity of NA-ergic and strengthening of activity of the DA-ergic and 5-HT-ergic brain systems. In other words, Wistar rats, differing in their degree of resistance to stress and characterized under normal conditions by normal biogenic amine levels in their brain structures, are also characterized by particular types of orienting-investigative behavior. This conclusion is confirmed by the work of other investigators [1, 2]. For instance, studies by Benešova showed that in rats with depressed investigative activity in an open field, and which she called "inactive rats," the NA level is low but the dopamine level high [2]. This is also supported by the results of investigations showing that the DA content in brain structures is raised, the 5-HT level is high [17], and their NA level is low [8] in rats with low motor activity, with which increased emotional reactivity is associated [9]. Our experiments also showed that R animals exhibit higher investigative activity than NR in an unfamiliar situation, and they have a higher NA level and lower DA and 5-HT levels in their brain. Meanwhile, NR rats, with a lower brain NA level, exhibit increased investigative activity in a burrow chamber. Similar results were obtained by other workers [12], who showed that the level of locomotor activity of rats with a low brain NA concentration is lower in a completely novel situation, and higher under conditions approximating to natural forms of life. It has also been shown that predominance of activity of the NA-ergic system correlates with a higher, and of the 5-HT-ergic system with a lower, level of all behavioral parameters of CNS activity [10]. It can thus be postulated on the basis of our own results and of data in the literature that an important role in the regulation of investigative behavior in animals differing in their resistance to stress was played by differences in the genetically determined inborn relationship between activity of the serotoninergic, dopaminergic, and noradrenergic systems of the brain.

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