Effect of Imipramine on Nerve Excitability in GC Rats

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Imipramine test (7.5 mg/kg) revealed a persistent positive reaction of Wistar rats, which manifested in reduced excitability of animals. Oral administration of imipramine solution was followed by unstable behavioral reactions in GC rats. Norepinephrine concentration in the cortical and limbic regions of these animals remained unchanged, while plasma corticosterone concentration decreased to the control level and did not differ from that in Wistar rats. Our results indicate that imipramine has a modulatory effect on destabilization of the adaptive system in catatonic GC rats.

Key Words: *catatonia; norepinephrine; serotonin; 5-hydroxyindoleacetic acid; corticosterone*

Catatonic GC rats is a genetic model for studying of complex "bipolar" behavior, including a variety of behavioral disorders from cataleptic freezing (akinetic pole) to increased excitability or nervousness (hyperkinetic pole) [2,3]. Hyperkinesia of this strain manifests in higher amplitude of the startle response [5], "irritable aggressiveness", and increased aggressive reaction in the glove test. GC rats exhibit a defense response to an unknown object, vocalization, jumps, and runs. The nature of hyperkinetic reactions remains unknown, because these animals were selected for passive-defense response of freezing. Previous studies showed that tricyclic antidepressant imipramine reduces freezing response in GC rats [4].

Here we studied the effect of imipramine on increased excitability (hyperkinetic pole), behavioral parameters, and neurohormonal state of catatonic GC rats.

MATERIALS AND METHODS

Experiments were performed on adult male GC rats (65th generation of selection, n=40) and Wistar rats (n=38). The animals were maintained in a vivarium under standard conditions (Institute of Cytology and Genetics) and had free access to water and food.

The animals were divided into 4 groups: control Wistar (n=28) and GC rats (n=30) and imipramine-treated (7.5 mg/kg orally, Sigma) Wistar (n=10) and GC rats (n=10). Test for "nervousness" and "bell ring test" were performed 3 times per month (according to selection criteria) and the results were scored.

The degree of "nervousness" was rated as follows:
- 0: no response to moving a stick over the cage bars;
- 1: startle response to moving a stick over the cage bars; and
- 2: jumps and running in response to moving a stick over the cage bars.

The results of the "ring test" were evaluated as follows:
- 0: no response;
- 0.2: head rotation;
- 0.5: whole-body rotation;
- 1: running over the width of the cage;
- 2: running over the length of the cage;
- 2.5: running over the diagonal of the cage; and
- 3: jumping out of the cage.

For measurement of monoamine concentrations in the brain and blood sampling, the animals were rapidly decapitated, the frontal cortex, hippocampus,

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hypothalamus, and adrenal glands were isolated and stored at -80°C until fluorometric assay of norepinephrine (NE), serotonin (5-hydroxytryptamine, 5-HT), and 5-hydroxyindoleacetic acid (5-HIAA) [1]. Monoamine fluorescence was measured on a Victor device (Perkin Elmeter) at 355/460 nm.

Plasma corticosterone concentration was measured by radioimmunoassay with [1,2,6,7-^{3}H]-corticosterone (Amersham) and Corticosterone antiserum (Sigma-Aldrich).

The significance of differences between the mean values was estimated by Student's *t* test and Newman—Keuls multiple comparison test.

RESULTS

Significant interstrain differences were found in the reaction of "nervousness" and length of "ring-induced runs" between intact animals ($p<0.001$, Fig. 1).

Administration of imipramine significantly decreased the "nervous" reaction and number of "ring-induced runs" in Wistar rats (3rd week of testing).

The "nervous" reaction and "ring response" in GC rats were significantly reduced after the 1st test. Interstrain differences in the control responses and "nervous" reaction were less pronounced, but remained significant in the 2nd test. However, the number and degree of runs in imipramine-treated GC rats were 2-fold higher than in control animals of the same strain. Two parameters of nervous excitability did not differ in animals of the control and treatment groups on the 3rd week of testing (Fig. 1).

Behavioral signs reflect a positive effect of the antidepressant imipramine, which relieves the symptoms of "nervousness" and reduces excitability in Wistar rats. Imipramine causes an unstable behavioral response of GC rats. This effect illustrates dysregulation of the adaptive system in catatonic GC rats.

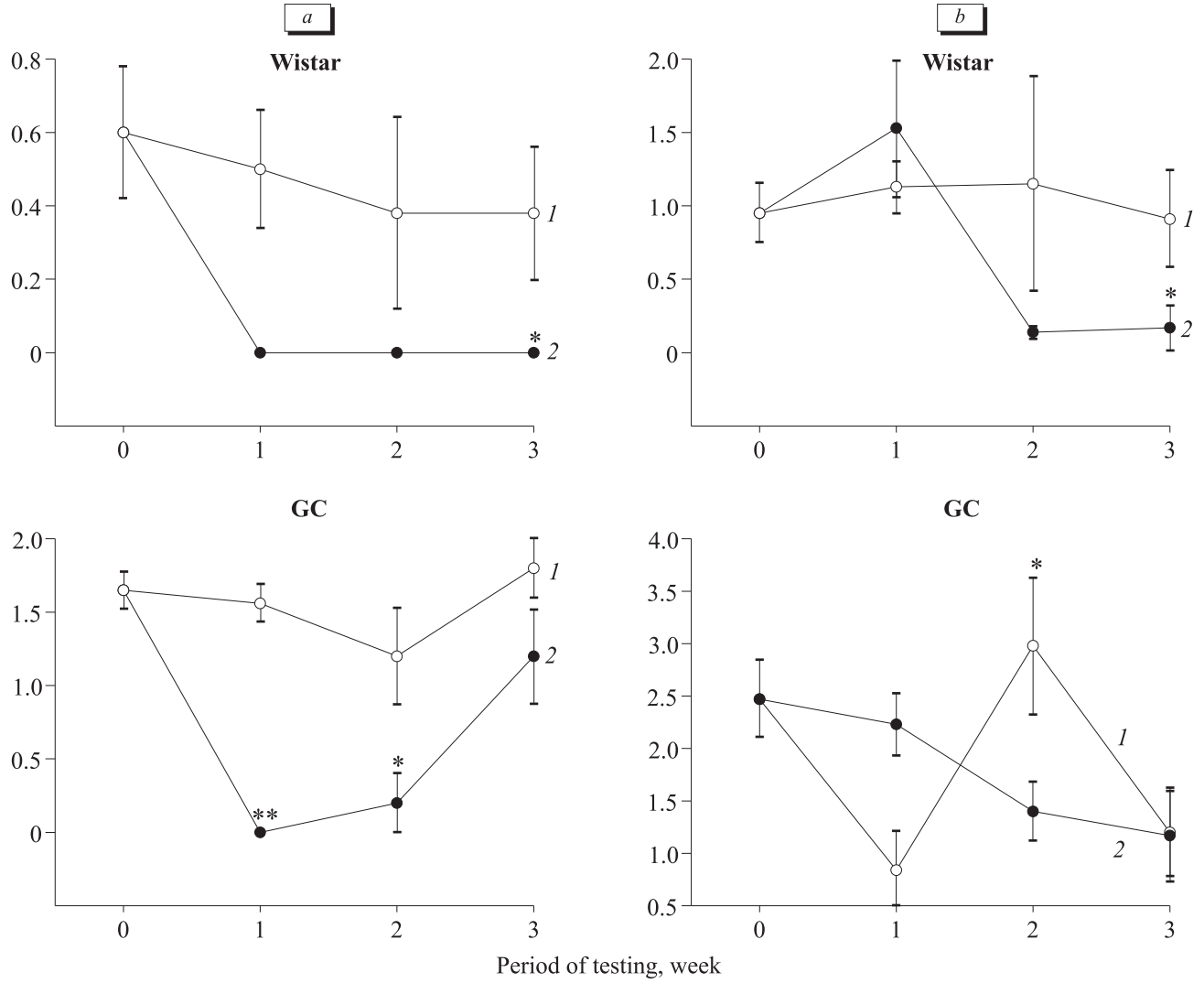

Fig. 1. "Nervous" excitability of Wistar and GC rats after administration of imipramine. "Nervousness", score (*a*); length of "ring-induced runs", score (*b*). Control (*1*) and imipramine (*2*). *$p<0.05$ and **$p<0.001$ compared to the control.

TABLE 1. Monoamine Concentration in Imipramine-Treated Wistar and GC Rats (μg/kg, *M*±*m*)

Test object	Group	NE	5-HT	5-HIAA
Frontal cortex	Wistar (control)	0.20±0.05 (8)	—	0.60±0.18 (8)
	Wistar (imipramine)	0.52±0.10 (7)**	—	0.40±0.07 (9)
	GC (control)	0.47±0.15 (6)	—	0.79±0.16 (9)
	GC (imipramine)	0.47±0.11 (7)	—	0.34±0.10 (9)*
Hypothalamus	Wistar (control)	1.17±0.17 (8)	0.62±0.39 (4)	0.38±0.06 (6)
	Wistar (imipramine)	1.15±0.18 (10)	1.27±0.33 (5)	0.29±0.06 (7)
	GC (control)	1.11±0.13 (10)	0.96±0.28 (5)	0.50±0.18 (10)
	GC (imipramine)	1.15±0.07 (10)	0.55±0.18 (4)	0.31±0.05 (9)
Hippocampus	Wistar (control)	0.25±0.02 (8)	0.46±0.12 (6)	0.26±0.06 (5)
	Wistar (imipramine)	0.21±0.01 (10)*	0.42±0.11 (8)	0.14±0.01 (10)*
	GC (control)	0.28±0.02 (10)	0.35±0.05 (10)	0.16±0.02 (8)
	GC (imipramine)	0.27±0.02 (9)	0.32±0.07 (7)	0.19±0.03 (9)
Adrenal glands	Wistar (control)	12.2±1.7 (8)	0.26±0.05 (5)	0.29±0.05 (8)
	Wistar (imipramine)	15.5±1.9 (10)	0.66±0.25 (5)	0.53±0.06 (10)**
	GC (control)	18.0±2.8 (10)	0.80±0.17 (6)$^{+}$	0.58±0.16 (10)
	GC (imipramine)	12.5±1.2 (10)	0.73±0.13 (7)	0.40±0.07 (10)

Note. *p<0.05 and **p<0.01 compared to the control; ^{+}p<0.05 compared to Wistar rats.

Evaluation of the neurotransmitter status showed that imipramine-induced reactivity of the noradrenergic system is observed in the frontal cortex and hippocampus of Wistar rats. By contrast, the concentration of NE in brain structures remained unchanged in GC rats (Table 1). Similar results were obtained in studying the effect of exogenous factors on animals of these strains. NE concentration was affected in Wistar rats, but did not change in GC animals. These differences are probably related to predisposition of GC rats to a specific reaction of freezing (at the behavioral and neurotransmitter levels) [1,6].

Variations of 5-HIAA concentration were observed in the frontal cortex and hippocampus of GC and Wistar rats, respectively. No intergroup differences were revealed in the hypothalamus of Wistar and GC rats. 5-HT concentration could not be estimated in the frontal cortex due to small weight of this structure.

Interstrain differences were found in the functions and imipramine-induced responses of the autonomic nervous system.

Plasma corticosterone concentration in GC rats was 2-fold higher than in Wistar rats (85.19±15.08 and 35.3±9.3 ng/ml, respectively). Administration of imipramine was followed by a decrease in plasma corticosterone concentration in GC rats, which did not differ from that in Wistar rats (36.23±13.16 and 32.59±7.06 ng/ml, respectively).

NE concentration in the adrenal medulla of GC rats was slightly higher than in Wistar rats. Interstrain differences were revealed in activity of the serotoninergic system in these glands (Table 1). 5-HT concentration in the adrenal medulla was elevated in GC rats.

Activation of the adrenal glands in intact GC rats was confirmed by variations in the following three parameters: concentrations of NE and 5-HT in the adrenal medulla; and plasma corticosterone level (*i.e.*, activity of the adrenal cortex). Activation of the adrenal glands probably contributes to increased excitability and high anxiety of GC rats. Treatment of Wistar rats with imipramine was also followed by an increase in the concentration of 5-HIAA, which serves as the major metabolite of the cerebral serotoninergic system.

The test for imipramine revealed persistent positive reaction of Wistar rats, which manifested in reduced excitability of animals. This treatment was followed by unstable behavioral response of GC rats. NE concentration in various brain structures of GC rats remained unchanged, while corticosterone concentration in these animals decreased by 2 times. Opposite changes in behavioral, transmitter, and hormonal parameters reflect dysfunction of the main regulatory systems in GC rats. Our results confirm the hypothesis that GC animals serve as a convenient model of depression [3,7,8].

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