# Effect of Low Doses of Piracetam on Conditioned-Response Memory in Rats

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Effects of low doses of piracetam, a psychotropic nootropic, on the memory of rats are studied. A positive effect of the drug in a dose many times lower than the doses used routinely is demonstrated on a model of elaboration of the active avoidance reaction.

**Key Words:** *piracetam; low doses; memory; conditioning*

Piracetam is the reference psychotropic drug belonging to the class of nootropics, agents which optimize the functions of the brain, notably memory [3]. Scientists working in various laboratories have experimentally (with rats) determined the doses of the drug which improve the processes of memory: 300 and 500 mg/kg [3,7].

The results of studies of the evolution of molecular mechanisms of memory have shed light on the positive effect of piracetam on memory. The modification of the brain's genome (induction of DNA synthesis) during learning has been experimentally proven.

The effects exerted by low doses of various chemicals, including neuropeptides, on the functional status of animals remain unclear [1,2,6], and, hence, a study of the effects of psychotropic agents in low doses on the higher brain functions seemed to be warranted.

Our purpose was to investigate the effect of low doses of piracetam on the time course of memory formation in rats.

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## MATERIALS AND METHODS

The study was carried out with 52 outbred rats weighing 180 to 200 g. Three experimental series were performed. In the first series 29 rats were used, divided into 3 groups. Group 1 ($n$=14) animals were injected piracetam in low doses ($1.6\times10^{-3}$ mg/kg), group 2 ($n$=6) the standard piracetam dose (500 mg/kg), and group 3 ($n$=9) normal saline. In the second series of experiments the animals were divided into 2 groups. Group 1 ($n$=8) received the drug in the aforesaid low dose, group 2 ($n$=7) normal saline. In the third series the animals were injected low doses of the drug ($n$=8) or normal saline ($n$=7). All the solutions were injected intraperitoneally in a volume of 0.5 ml 30 min before the beginning of the experiment. Solutions of low piracetam doses were prepared as follows. The necessary quantity of the drug was weighed with an accuracy of up to $10^{-5}$ g in thin-walled weighing bottles. Then the dry agent in the weighing bottle was put in a measuring flask with the solution. After thorough stirring the solution was poured into a 1- or 2-liter measuring flask and the volume brought to the needed quantity. The calculations were carried out in mol/liter of solution (for example, dry agent for 1.41 mg/liter of solution is $10^{-5}$ mol/liter). Each concentration was prepared separately, without resorting to diluting ready-made solutions.

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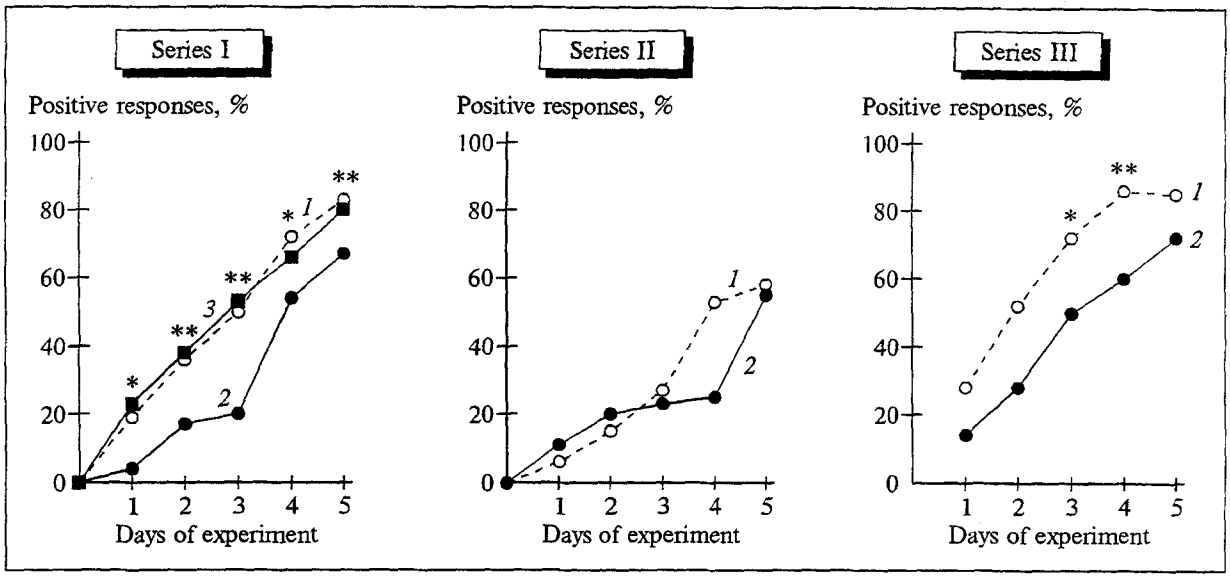


**Fig. 1.** Effect of different doses of piracetam on the avoidance reaction in rats. *1*) low doses ($1.6\times10^{-3}$ mg/kg); *2*) control (normal saline); *3*) standard doses (500 mg/kg). $^{*}p<0.05$, $^{**}p<0.01$ in comparison with the control.

The presence of a true solution (characterized by pharmacologic action) was monitored by high-performance liquid chromatography on an HPZC (Beckman) chromatographer using an Ultrasphera C18 ODC 25×4.6 mm column.

Electrodefense conditioned responses, elaborated in a shuttle box over 5 days, served as the model of memory. The experimental protocol we used yielded an average level of 75 to 85% positive reactions (avoidance reaction in response to the conditioned stimulus). Experiments were carried out every other day using the following protocol: the conditioned stimulus (800 Hz sound) was switched on and, after 10 sec, an electric current. If the animals ran to the other part of the box, both stimuli were switched off; if they stayed where they were, they were shocked. Thus, conditioning was achieved. The results of the experiments were processed using Wilcoxon's and Kolmogorov-Smirnov's tests.

## RESULTS

The experimental data indicate that in both high and low doses piracetam appreciably accelerated the formation of the avoidance reaction in rats of the first and third series (Fig. 1). The magnitude of the avoidance reaction of rats administered piracetam in the second series of experiments surpassed that of control animals only on day 4, but this difference was statistically unreliable ($p=0.06$, Fig. 1). These ambiguous results correlate well with the reports according to which the drug sometimes accelerates learning, and sometimes does not [3,4].

Two-factor analysis showed that, starting from day 1, low doses of the drug exerted a statistically reliable effect on the process of elaboration of the avoidance reaction ($p<0.001$, Fig. 1). Hence, the effect of piracetam in a dose of $1.6\times10^{-3}$ mg/kg on the formation of the avoidance reaction is comparable to its effect in a dose of 500 mg/kg.

We do not yet quite understand the mechanism behind the efficacy of low doses. The possibility of individual molecules meeting target cells, as Ashmarin has proposed [1], should not be ruled out.

Piracetam in a dose improving the memory processes is known to activate the rat brain genome [9]. A similar mechanism of action of low doses of the drug on memory may be hypothesized, and in this case we must acknowledge the possibility of individual molecules of piracetam meeting individual genes.

These facts call for further experiments involving a detailed analysis of the effects of low doses of not only piracetam, but of other psychotropic agents as well. The detected regularities are interesting in themselves, as evidence of a positive effect on memory of a psychotropic agent in doses much lower than those routinely used.

The results of these experiments are important for exploring the concept of a universal pattern of macromolecular mechanisms whereby psychotropic agents of different chemical nature act upon the higher brain functions [8].

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## REFERENCES

1. I. P. Ashmarin, T. V. Lelekova, and L. Ts. Sanzhieva, *Izv. Akad. Nauk SSSR, Ser. Biologiya,* № 4, 531-536 (1992).

2. E. B. Burlakova, A. A. Konradov, and I. V. Khudyakov, *Ibid.*, № 2, 184-194 (1990).
3. T. A. Voronina, in: *Pharmacology of Nootropics (Experimental and Clinical Studies)* [in Russian], Moscow (1989), pp. 8-19.
4. A. N. Inozemtsev and L. L. Pragina, *Vestn. Mosk. Universiteta,* Ser. *Biologiya,* № 4, 23-31 (1992).
5. T. V. Lelekova, P. N. Aleksandrov, P. Ya. Romanovskii, and I. P. Ashmarin, *Byull. Eksp. Biol. Med.,* **108**, № 7, 8-10 (1989).
6. T. V. Lelekova, I. P. Ashmarin, and P. N. Aleksandrov, *Vestn. Mosk. Universiteta, Ser. Biologiya,* № 3, 14-18 (1992).
7. L. L. Pragina, T. A. Voronina, A. N. Inozemtsev, *et al., Farmakol. Toksikol.,* **53**, № 3, 8-9 (1990).
8. N. A. Tushmalova, *Zh. Vyssh. Nervn Deyat.,* **44**, № 1, 3-7 (1994).
9. N. A. Tushmalova, V. G. Bezlepkin, F. F. Kokaeva, and A. I. Gaziev, *Dokl. Akad. Nauk SSSR,* **320**, 761-763 (1991).

# Study of the Hemostatic Properties of Gaseous Ozone

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The hemostatic properties of ozone were studied on models of parenchymatous (from a wound in the liver) and stem (from the stump of the tail) bleedings in rats. An air-ozone mixture at a flow rate of 1 liter/min and a concentration of 2 mg/ml was found to exert a pronounced hemostatic effect. Our findings indicate that the arrest of bleeding under the influence of ozone is due to the formation of a fibrin membrane on the surface of the flowing blood, this leading to rapid and effective hemostasis. Preliminary drying of the wound still further speeds the onset of hemostasis.

**Key Words:** *gaseous ozone; parenchymatous bleeding; stem bleeding; hemostasis; rats*

Despite the ever-growing arsenal of medicamental (matrix) agents for local hemostasis [1], remote methods of arresting bleeding are still preferred in abdominal surgerry, methods that are based on the use of physical factors, such as cryo, thermo-, aerothermo-, laser, and plasma coagulation. However, hemostasis created by means of the majority of these methods involves the formation of a film of necrotic tissue of varying thickness on the surface of the wound, and this may adversely affect subsequent healing [2,6,9].

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For this reason, the influence of other, less damaging products, appears to be interesting, gaseous ozone being one of them [3]. Ozone, an allotropic modification of oxygen, is present in large quantities in the upper layer of the Earth's atmosphere [7] and forms naturally in stormy air and during some physicochemical processes [4,5]. The medical use of ozone is predicated upon its antibacterial and oxidative characteristics. It is used in the treatment of wounds and burns, in detoxication therapy, for the decontamination of water, and for the sterilization of medical instruments. Its immunomodulating, antiinflammatory, and antistress properties are also being studied. Published reports about the effects of ozone on whole blood provide information only about the correction of anemia and normalization of blood rheology during artificial circulation.

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