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The Influence of Bromocriptine on the Sexual Activity in Ethanol-exposed Male Rats

Wpływ bromokryptyny na aktywność płciową szczurów poddawanych wielokrotnemu działaniu etanolu

Влияние бромкриптина на половую активность крыс получающих этанол

Long-lasting intake of ethanol and its influence on certain changes in the activity of male sexual hormones is the subject of numerous experiments (13). It is suggested that the decreased libido, hypothrophy of testicles and impotence in individuals subjected to ethanol, are connected with the lowered level of testosterone.

Various mechanisms may be responsible for such an activity of ethanol. There has been pointed out that long-lasting intake of ethanol quickens the metabolism of testosterone in the liver (15). The results of other experiments indicate that ethanol and/or its metabolite, acetaldehyde decrease the biosynthesis of testosterone in the Leidig cells (9). It has been proved, too, that ethanol reduces the number of testicular gonadotropin receptors what, aditionally, decreases the synthesis of testosterone (2).

Another reason for the disordered function of gonads in alcoholics may be the influence of ethanol on the central nervous system structures. Particularly, it refers to the neurones of hypothalamus because they can regulate excretion of the hormones of anterior pituitary gland. In this way, hypothalamus may affect not only the level of the hormones produced by gonades but also to release pituitary prolactin.

The present state of research indicates that ethanol results in clear changes in the function of the dopaminergic system of the brain. The influence of ethanol upon the neurones of this system depends on its dose and the period of activity. Most researches point out that prolonged administration of ethanol leads to hypofunction of dopaminergic neurones of the brain (7, 8).

The data have been used as an inspiration to carry out the research on the sexual activity of rats exposed to multiple effect of ethanol. In these animals there have been estimated their sexual activity as well as the influence of bromocriptine on sexual responses.

METHODS

The experiments were completed on white male Wistar rats weighing 250—300 g. The place in which they were kept was lit with electricity from 6 p.m. till 6 a.m. For the remaining 12 hrs the rats stayed in darkness. All the animals were kept in separate cages and had free access to water and food (rodent pellets, Bacutil).

The investigations were carried out on 22 rats. The group of 14 rats received i.p. p-chlorophenylalanine (PCPA) and ethanol intragastrically. The second group of 8 rats was given i.p. PCPA and water instead of ethanol. Injections of PCPA dosed 100 mg/kg were applied at 7 p.m. on the first, the third, and the fifth day of the experiment duration. Ethanol was administered as the 40 vol. % water solution dosed 3 g/kg, every 12 hrs for 5 days. On the fourth and the fifth day of the experiment the actual dosage of ethanol was increased by 1 g/kg. The last dose of ethanol and PCPA was administered at 7 p.m. on the 5 day of the experiment.

The sexual activity of the male rats was assessed twice: 14 and then 24 hrs after withdrawing ethanol and PCPA. Before the observation started the animals with ethanol and PCPA had been divided into 2 groups consisting of 7 rats. In one of these groups the rats received i.p. Bromocriptine mesylate (Sandoz, Basel) in doses of 2.5 mg/kg 30 min. before each observation. The rats in the remaining groups received i.p. saline.

5 min. before the observation the male rats were separately placed in open cages of $50 \times 50 \times 50$ (cm). After a five-minute period of adaptation the female rats were put into the cages and for the following 30 min. the male rats' sexual activity was observed with the assessment of such episodes as getting interested in the in the sexual organs of female rats, covering females without copulative movements, and with these movements as well.

The obtained results were presented in the form of average rate $\pm$ the standard error. Statistical analysis was carried out using Student's *t*-test.

RESULTS

The experiments were carried out on rats which were treated with PCPA to increase their sexual activity. It was proved that multiple application of ethanol decreased clearly sexual activity which was otherwise raised by PCPA. The decreased sexual activity lasted for more than 24 hrs since the withdrawal of ethanol.

Bromocriptine (agonist of dopaminergic receptors) was administered to those rats which had been receiving PCPA and ethanol. Injections of bromocriptine were applied twice: after 14 and 24 hrs since removing ethanol. The administration of bromocriptine after 14 hrs helped only slightly to increase the sexual activity of the rats. During 30 min. observation the number of copulative movements with ejaculation increased from 5.71 ± 0.81 to 7.88 ± 1.13 (Table 1). After 24 hrs since the withdrawal of ethanol, the animals received bromocriptine again. There was observed then a statistically significant increase in the sexual activity of the male rats. Bromocriptine caused more than quadruple increase in the number of copulative movements with ejaculation (Table 2). The remaining parameters used in estimation of the male rats' sexual activity did not undergo any significant changes.

Table 1. The influence of bromocriptine on the sexual behaviour in the male rats exposed to ethanol

Drugs	Sexual behavior		
	I	II	III
PCPA	12.43±0.9	12.86±1.13	14.0 ±1.45
PCPA+E	1.29±0.68**	7.29±0.79**	5.71±0.81**
PCPA+E+Bc	3.29±0.48	6.0 ±0.87	7.88±1.13

The rats received bromocriptine in dose 2.5 mg/kg 14 hrs after withdrawal of ethanol.

Explanation: PCPA — p-chlorphenylalanine, E — ethanol, Bc — bromocriptine, I — interest in the sexual organs of female rats, II — covering females without the copulative movements, III — covering females with copulative movements. Data are shown as mean±SE.

** $p<0.005$ — statistical results are significant in comparison to PCPA.

Table 2. The influence of bromocriptine on the sexual behaviour in the male rats exposed to ethanol

Drugs	Sexual behavior		
	I	II	III
PCPA	14.0±1.30	6.9±1.13	24.8±1.82
PCPA+E	1.7±0.68**	2.0±0.79**	2.3±0.89*
PCPA+E+Bc	1.9±0.59	3.4±0.90	9.9±1.20***

The rats received bromocriptine in dose 2.5 mg/kg 24 hrs after withdrawal ethanol.

* $p<0.05$; ** $p<0.005$ — statistical results are significant in comparison to PCPA.

*** $p<0.005$ — statistical results is significant in comparison to PCPA+E.

Explanation see Table 1.

DISCUSSION

Compounds decreasing the activity of the serotoninergic system, for example, PCPA, intensify sexual responses in male rats. In the experiments described above, strong sexual activity of male rats was reached by i.p. administering of PCPA, in a dose of 100 mg/kg, on the first, the third, and the fifth day of ethanol application. The results of these experiments indicate that multiple application of ethanol decreased sexual reactions in rats receiving PCPA. The period of thus lowered sexual activity in male rats lasted longer than 24 hrs. Taking into consideration the fact, that PCPA decreases the serotoninergic system activity due to lowered synthesis of serotonine (11), it can be hinted that the disordered sexual activity in male rats, caused by ethanol, may be the result of its influence on other neurotransmitting system of the brain.

It is concluded, from the so far carried out experiments, that ethanol administered protractedly modifies central neurotransmission, although some contradictory results have also been obtained (7). Changes in the activity of some neurotransmitting sytems in the brain, have been confirmed by behavioral experiments on animal and by clinical examination of alcoholics. It has been proved, among others on animals, that dopamine administered into the brain restrains the symptoms of ethanol abstinence (4), and haloperidol — a blocker of central receptors sensitive to dopamine — increased seizures as the symptom of ethanol withdrawal in mice (3). Besides, Alkana et al. (1) have found that L-DOPA may decrease the disorders of motor coordination and divisibility of attention in people, caused by ethanol. The influence of ethanol on the activity of the central dopaminergic system was investigated by Lai et al. (12) as well. Their results indicate that long-lasting administration of ethanol increases the number of places binding with the ^{3}H-spiroperidol in the striatum of the brain. Stereotypy caused by apomorphine was stronger, too, in rats receiving ethanol than in the control group. Experiments with the use of bromocriptine supply additional evidence for participation of the dopaminergic system in obtaining the results of long-lasting administration of ethanol. It has been found that this substance restrains the symptoms of ethanol withdrawal in the experimental rats (6), as well as in alcoholics (5). Considering the fact that bromocriptine increases the activity of the neurones of the dopaminergic central nerve system, it is suggested that multiple administration of ethanol results in the disfunction of these neurones.

Other pharmacological studies on the activity of bromocriptine indicate that this substance has a complex mechanism of its impact. Bromocriptine may work not only as an agonist of the dopaminergic receptors D_2 but also as an antagonist of the dopaminergic receptors D_1 (10). Its activity depends, to a large extent, on an administered dose.

In the performed experiments bromocriptine was twice administered in doses 2.5 mg/kg each. Statistically significant activity of bromocriptine occurred after the second dose, which was injected after 24 hrs, counting from the last administration of ethanol. It was observed then a significant increase in the number of copulative movements with ejaculation in rats treated with ethanol. This symptom is the basic one in the behavioral estimation of the male rats sexual activity. The increased number of sexual reactions due to bromocriptine indicates that long-lasting administration of ethanol could evoke certain changes in the central dopaminergic system of male rats. Simultaneously, these experiments proved the existence of some connection between the appearance of the decreased sexual activity in rats receiving ethanol protractedly and the function of the central dopaminergic neurones.

It seems that the research on the influence of ethanol on the amount of prolactine in the blood of alcoholics or experimental animals might be

significantly related to the results of performed observation. Van Thiel et al. (18), as well as Majumdar (14), found that the amount of prolactine proved to be larger in the blood of alkoholics. Experiments carried out on rats supplied similar results (3). In male individuals, hyperprolactinaemia may be accompanied by the loss of libido, as well as by impotence and the symptoms of feminization. It seems then, that prolactine may be one of the factors responsible for the decreased sexual activity of rats undergoing the long-lasting impact of ethanol.

The mechanism of the appearance of hyperprolactinaemia caused by ethanol, may have connection with the activity of the dopaminergic neurones of the brain. It has been concluded, due to numerous experiments, that dopamine performs the function of the hypothalamus factor restraining secretion of prolactine (15). Thus, the ethanol evoked changes in the activity of the dopaminergic neurones in the brain may influence the amount of prolactine released from the anterior pituitary gland. The explanation of this problem requires carrying out further research.

REFERENCES

1. Alkana R. L. et al.: Reversal of Alcohol Intoxication in Humans: an Assessment of the Efficacy of L-dopa, Aminophylline and Ephedrine. Psychopharmacology **55**, 203, 1977.
2. Bhalla V. K., Chen C. J. H., Gnanaprakasam M. S.: Effects of *in vivo* Administration of Human Chorionic Gonadotropin and Ethanol on the Process of Testicular Receptor Depletion and Replenishment. Life Sci. **24**, 1315, 1979.
3. Blum K. et al.: Enhancement of Alcohol Withdrawal Convulsions in Mice by Haloperidol. Clin. Toxicol. **9**, 427, 1976.
4. Blum K. et al.: Suppression of Ethanol Withdrawal by Dopamine. Experienta **32**, 493, 1976.
5. Borg V., Weinholdt T.: Bromocriptine in the Treatment of the Alcohol Withdrawal Syndrome. Acta Psych. Scand. **65**, 101, 1982.
6. Gonzales Perez J. N., Gonzales-Quevedo A.: Blocade Audiogenic Seizures by Bromcriptine. Pharmacol. Biochem. Behav. **12**, 823, 1980.
7. Hunt W. A., Majchrowicz E.: Alterations in Neurotransmitter Function after Acute and Chronic Treatment with Ethanol. [in:] Biochemistry and Pharmacology of Ethanol. Vol. **2**, Majchrowicz E., Noble E. P. (eds.), Plenum Press, New York 1979.
8. Hunt W. A., Majchrowicz E.: Studies of Neurotransmitter Interaction after Acute and Chronic Ethanol Administration. Pharmacol. Biochem. Behav. **18**, Suppl. 1, 371, 1983.
9. Johnston D. E. et al.: Inhibition of Testosterone Synthesis by Ethanol and Acetaldehyde. Biochem. Pharmacol. **30**, 1827, 1981.
10. Kaiser C.: Structure-activity Relationships of Dopamine Receptor Agonists. [in] Dopamine Receptor Agonists. Poste G., Crooke S. T. (eds.), Plenum Press, New York–London 1984.
11. Koe B. K., Wiessmann A.: p-Chlorophenylalanine: a Specific Depletor of Brain Serotonin. J. Pharmacol. Exp. Ther. **154**, 499, 1966.
12. Lai H., Carino M. A., Horita A.: Effects of Ethanol on Central Dopamine Functions. Life Sci. **27**, 299, 1980.
13. Lindholm J. et al.: Pituitary-testicular Function in Patients with Chronic Alcoholism. Eur. J. Clin. Invest. **8**, 269, 1978.

14. Majumdar S. K.: Serum Prolactin Concentrations during the Hangover Phase of Ethanol Withdrawal Syndrome. Neuroendocrinol. Lett. **4**, 253, 1982.
15. Neil J. D.: Neuroendocrine Regulation of Prolactin Secretion. [in:] Frontiers in Neuroendocrinology. Martini L., Ganong W. F. (eds.), Raven Press, New York 1980.
16. Rubin E. et al.: Prolonged Ethanol Consumption Increases Testosterone Metabolism in the Liver. Science **91**, 563, 1976.
17. Seilicovich A. et al.: The Effect of Acute and Chronic Ethanol Administration on Prolactin Secretion in Male Rats. J. Androl. **3**, 344, 1982.
18. Van Thiel D. H., Gavaler J. S., Lester R.: Plasma Estrone, Prolactin, Neurophysin, and Sex Steroid Binding Globulin in Chronic Alcoholic Men. Metabolism **24**, 1015, 1975.

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STRESZCZENIE

Szczurom, samcom szczepu Wistar, podawano dożołądkowo 40% v/v wodny roztwór etanolu co 12 godz. w ciągu 5 kolejnych dni w dawce jednorazowej 3 g/kg. Równocześnie zwierzęta te otrzymywały p-chlorofenyloalaninę (PCPA) w dawce 100 mg/kg i.p. Aktywność płciową samców oceniano po 14 i 24 godz. od podania ostatniej dawki etanolu. Stwierdzono, że etanol zmniejszał aktywność płciową badanych zwierząt. Bromokryptyna — agonista receptorów dopaminergicznych — zastosowana w dawce 2,5 mg/kg i.p. odwracała tłumioną przez etanol aktywność płciową samców. Sugeruje się, że jedną z przyczyn zmniejszonej przez etanol aktywności płciowej samców może być dysfunkcja neuronów dopaminergicznych mózgu.

РЕЗЮМЕ

Самцам крыс в течение 5 дней с помощью интрагастральных интубаций подавали 40% v/v раствор этанола в дозах 3 г/кг с 12 часовыми интервалами. Одновременно эти животные получали п-хлорфенилаланин (PCPA) в дозах 100 мг/кг подаваемых внутрибрюшинно. Половую активность оценивали через 14 часов и 24 часа после введения этанола. Доказано, что этанол понижал половую активность исследуемых животных. Бромкриптин — агонист допаминэргических рецепторов, применяемый в дозах 2,5 мг/кг внутрибрюшинно восстанавливал подавленную этанолом половую активность самцов. Предполагается, что одной из причин пониженной этанолом половой активности самцов может быть дисфункция допаминэргических нейронов мозга.