EFFECT OF LITHIUM CHLORIDE ON ETHANOL CONSUMPTION BY RATS

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Selective consumption of ethanol was produced in rats in the course of 2 months by giving a 5% solution of ethanol as the sole source of fluid. Lithium chloride, given by intraperitoneal injection in a dose of 35 mg/kg twice a day for 14 days, depressed the preference for ethanol by producing reversal of motivations, the mechanism of which is connected with changes in the activity of the hypothalamic centers of neuroendocrine regulation. The possible use of lithium salts in the treatment of chronic alcoholism is discussed.

KEY WORDS: lithium; ethanol consumption; hypothalamic neurosecretion.

Among the remedies provided by modern psychopharmacology lithium salts have gained general acceptance for the prevention and treatment of endogenous affective psychoses [1]. The first attempts have been made to use lithium salts for the treatment of various types of drug addiction [7, 10]. Emotional disorders are known to be the most characteristic clinical manifestations of chronic alcoholism [5]. These facts justify the experimental study of the effect of lithium salts on the course of alcohol addiction. The mechanism of action of alcohol and the pathogenesis of chronic alcoholism are linked with disturbances of the activity of the motivation and adaptation centers of the hypothalamus [6]. The hypothalamo-hypophyseal neurosecretory system (HHNS) is an extremely important structural and functional divison of the hypothalamus, through which a wide range of regulatory influences of the CNS on numerous functions of the body are exerted [3].

The study of the state of the HHNS during the treatment of experimental chronic alcoholism is thus an important step in the elucidation of the possible mechanism of the psychotropic action of lithium salts and the establishment of the pathogenesis of this disease.

EXPERIMENTAL METHOD

Noninbred male albino rats weighing initially 120-150 g, in which preference for ethanol had been formed in the course of 2 months by providing a 5% solution of ethanol as the sole source of fluid, were used as the model of chronic alcoholism. At the end of this period the rats were given free access to fluid (5% ethanol solution or water) only in the experimental cages during the 3 h of the experiment. Lithium chloride, in close to the therapeutic dose (35 mg/kg) was injected as a 5% solution intraperitoneally twice a day for 14 days: 30 min before the animals were placed in the experimental cages and 8 h after the first injection. Control animals received the same volume of distilled water under the same conditions. At the end of the experiment the experimental and intact animals were decapitated and the hypothalamus and pituitary were fixed in Bouin's or Carnoy's fluid. Serial paraffin sections of these structures were stained as follows: with paraldehydefuchsin by the Gomori–Gabe method with counterstaining by Halmi's mixture to demonstrate neurosecretion, and by gallocyanin by Einarson's method to demonstrate nucleic acid (RNA). The morphological and functional state of the supraoptic (SON) and paraventricular (PVN) nuclei of the hypothalamus was assessed on the basis of the relative numbers of the different types of neurosecretory neurons [3] and their RNA content, followed by calculations of the mean histochemical coefficient [4]. The content of neurosecretion in the median eminence (ME) and the principal posterior lobe (PPL) of the neurohypophysis was estimated by a five-point system [3].

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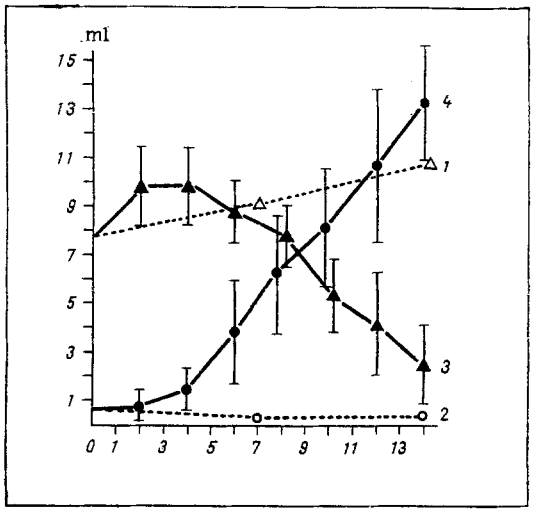

Fig. 1

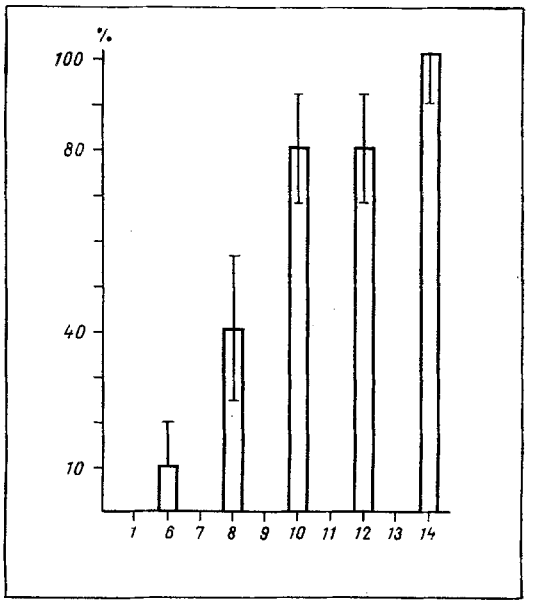

Fig. 2

Fig. 1. Effect of course of lithium chloride injections (35 mg/kg, twice a day fc 14 days) on consumption of water and 5% ethanol solution in rats with preestablished selective preference for ethanol. Control: 1) ethanol consumption; 2) water consumption; experiment: 3) ethanol consumption; 4) water consumption. Abscissa, days of injection of compound; ordinate, quantity of fluid per animal (in ml).

Fig. 2. Number of rats with reversal of motivation (in %) during course of injections of lithium chloride (explanation in text).

EXPERIMENTAL RESULTS

The model of chronic alcoholism chosen is sufficiently adequate, for at the end of 2 months of the experiment, under conditions of free choice, all the rats preferred the ethanol solution to water. Lithium chloride had a marked effect on the character of the relationship formed (Fig. 1). Starting from the sixth day of administration of the compound an increase in the water consumption was observed, whereas there was no change in the ethanol consumption during this period of observation and the significant decrease in ethanol consumption occurred only after administration of lithium salt for 10 days. At the end of the 14-day course of lithium chloride a marked increase in the water consumption and a decrease in the ethanol consumption were observed ($P = 0.001$). In the animals of the control group no significant changes were found in the water and ethanol consumption throughout the period of observation. Under the influence of lithium chloride reversal of motivation, i.e., restoration of preference for water in the total volume of liquid consumed, developed in all the animals by the 14th day (Fig. 2).

Considerable changes also were found in a study of the morphological and functional state of the HHNS of the experimental animals. In rats with preference for ethanol clear signs of inhibition of the HHNS were recorded. In the neurosecretory nuclei of the hypothalamus the number of actively functioning type 1a neurons was sharply reduced and the number of neurons with low activity (types 1c and 2) and of pycnomorphic cells (type 3) was increased (Table 1). Inhibition of the synthesis of neurohormones in SON and PVN also is confirmed by the significant ($P = 0.001$) decrease in the RNA content in their neurons to 1.61 ± 0.07 conventional units (2.32 ± 0.04 conventional units in the intact animal) and 1.68 ± 0.09 conventional units (2.33 ± 0.05 conventional units in the intact animal). Besides inhibition of synthesis of the neurosecretion, its transport along the hypothalamo-hypophyseal tract also was considerably inhibited, as shown by the appearance of numerous fragments of neurosecretory fibers, the large and giant expansions of which were packed with homogeneous masses of neurosecretion. The ME and PPL of these animals were indistinguishable in their content of neurosecretion from ME and PPL of the intact rats, but the considerable number of Herring's bodies and the reduction of the capillary network of these formations are evidence of a decrease in the secretion of neurohormones into the bloodstream.

TABLE 1. Effect of Course of Intraperitoneal Injections of Lithium Chloride (35 mg/kg twice a day for 14 days) on Number (in %) of Different Types of Hypothalamic Neurosecretory Neurons in Rats with Preestablished Selective Preference for Ethanol (mean of five observations, M ± m)

Type of neuron	Intact animals	Experimental animals								
		before injection of LiCl (2)	P_{1-2}	injection of water (control; 3)	P_{1-3}	P_{2-3}	before injection of LiCl (2)	P_{1-4}	P_{2-4}	P_{3-4}
Supraoptic nucleus										
1a	64±5,5	18±2,1	0,001	25±3,0	0,001	0,1	80±3,2	0,1	0,001	0,001
1b	27±5,1	47±1,2	0,01	41±2,7	0,05	0,1	5±1,5	0,001	0,001	0,001
1c	5±1,0	20±1,9	0,001	21±1,9	0,001	0,5	—			
2	1±0,6	4±1,2	0,05	5±1,5	0,05	0,5	—			
3	3±0,2	11±1,0	0,001	8±1,5	0,01	0,2	15±3,2	0,001	0,5	0,1
Paraventricular nucleus										
1a	34±1,7	9±1,9	0,001	15±3,2	0,001	0,1	39±3,6	0,2	0,001	0,01
1b	47±2,1	28±4,0	0,01	32±5,7	0,05	0,5	45±4,5	0,5	0,02	0,5
1c	11±2,5	40±4,0	0,002	36±3,6	0,002	0,5	9±1,7	0,5	0,001	0,001
2	4±0,8	16±2,1	0,001	12±1,7	0,001	0,1	2±0,6	0,1	0,001	0,001
3	4±0,2	7±0,2	0,01	5±0,8	0,5	0,1	5±1,0	0,1	0,2	0,5

After injection of water into animals with preference for ethanol (control group) the character of the response of the HHNS showed no significant change. However, the increase in the RNA content in the SON and PVN neurons (by 6-8%), as well as the decrease in the quantity of neurosecretion deposited along the course of the neurosecretory fibers and in their endings, coupled with the moderate dilatation of the vascular network of the neurohypophysis, suggests some activation of the neurosecretion process.

Lithium chloride significantly changed the morphological and functional state of the HHNS in rats with preference for ethanol consumption. In the animals of this series the predominant elements in the morphological picture of SON were large cells, with indistinct boundaries of their bodies and nuclei, and with diffusely vacuolated cytoplasm, containing no neurosecretion and little RNA (1.89 ± 0.04 conventional units; $P = 0.001$). Neurosecretory fibers containing granules of neurosecretion were not found on the territory of SON or along the course of the hypothalamo-hypophyseal tract. The quantity of neurosecretion in PPL was sharply reduced (by 70%) and the capillary network of this formation was dilated and congested with blood. The results described above are evidence of considerable activation of the neurosecretory process in the SON–PPL system. The morphological and functional characteristics of PVN and ME under these conditions were close to those of intact animals.

It follows from the facts described above that, by the character of their action on the HHNS, ethanol and lithium chloride are antagonists, for a course of injections of the latter compound leads to activation of the processes of synthesis, transport, and secretion of neurohormones previously inhibited by ethanol. It can be postulated that the changes found in the consumption of water are due to disturbances of the activity of the SON–PPL–antidiuretic hormone system.

Lithium chloride thus has the property of depressing preference for ethanol in rats, by causing reversal of motivations, the mechanism of which is probably connected with changes in the activity of the hypothalamic centers of neuroendocrine regulation. In the character of the above effect lithium chloride differs significantly from the other psychotropic agents whose influence on ethanol consumption has been studied previously [2]. Considering that lithium salts have the ability to prevent attacks of emotional disorders [9] and that their prolonged administration does not cause habituation [8], their use for the treatment of patients with chronic alcoholism would appear to be promising

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EFFECT OF PSYCHOTROPIC DRUGS ON DEFENSIVE CONDITIONING IN EXPERIMENTAL NEUROSIS

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Rats exposed to the prolonged action of an emotionally adverse factor (the response of a neighboring rat to nociceptive stimulation) develop neurosis as a result of which the course of defensive motor conditioning is disturbed. Benactyzine, diazepam, chlordiazepoxide, and sodium hydroxybutyrate improve the formation of defensive conditioned reflexes in neurosis of this type. Trioxazine, trifluoperazine, and amphetamine have no such action. The suggested model of experimental neurosis can be used for the study of antineurotic activity of psychotropic drugs.
KEY WORDS: neurosis; conditioned reflex; psychotropic drugs.

Tranquilizers are widely used to relieve emotional stress and to treat neurotic states. However, despite their broad common spectrum of psychotropic activity, not all tranquilizers have proved equally effective in the treatment of neuroses [1]. It is therefore important to be able to evaluate tranquilizers experimentally in order to discover and predict their therapeutic effect in neurotic states.

The object of this investigation was to study the effect of psychotropic drugs with tranquilizing properties on one form of experimental neurosis in rats.

EXPERIMENTAL METHODS

Experiments were carried out on 160 male albino rats weighing 250-350 g. The methods of the avoidance response to stimulation of the partner [5, 9] and formation of a motor defensive conditioned reflex [10] were used. The defensive conditioned reflex served as an indicator of the functional state of the CNS. Animals with a stable avoidance response to stimulation of the partner, observed not less than 10 times in succession, were chosen for the experiments. By this time the rats showed changes in behavior when put inside the chamber: a posture of alertness, sometimes accompanied by a squeak, resistance to being placed in the chamber, and jumping out of it. At the same time, autonomic somatic disorders appeared (baldness, keratitis). These phenomena form part of the picture of an experimental neurosis [7]. Experiments were carried out by the following scheme: The rat was placed for 5 min in the chamber for the avoidance response to stimulation of the partner, and then immediately transferred to the defensive conditioned reflex chamber. The conditioning stimulus was the ringing of a bell. It acted alone for 5 sec, after which an electric current was applied to the floor of a chamber (also for 5 sec) while the bell continued to ring. The rat was taught to jump onto a vertical rod and to support itself on it until the stimulus ceased to act. The intervals between stimuli were 1 min. Training continued for 10 days with daily presentation of 10 combinations. Rats not taught the avoidance response during stimulation of the partner served as the control.

Benactyzine, diazepam, chlordiazepoxide, trioxazine, sodium hydroxybutyrate, and trifluoperazine were used. Each substance was injected intraperitoneally into 10 rats with a neurosis and 10 control rats 30 min before the beginning of each training session, in daily doses, each of which causes inhibition of the avoidance response to stimulation of the partner (tranquilizing effect). Isotonic sodium chloride solution was injected into 10 rats with neurosis and 10 control rats. The number of jumps on to the rod in response to the conditioning

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