3. N. I. Maisov, T. D. Baimanov, and Yu. V Burov, Byull. Éksp. Biol. Med., No. 8 (1991).
4. T. G. Pukhal'skaya, N. I. Maisov, and R. S. Mirzoyan, Farmakol. Toksikol., No. 6, 39 (1989).
5. N. J. G. Barnes, A. J. Bradbury, B. Costall, et al., J. Nueral. Transmiss., Suppl. 20, 47 (1986).
6. T. Di Paolo, P. Bedard, M. Daigle, and R. Boucher, Brain Res., **379**, No. 2, 286 (1986).
7. R. W. Fuller and S. K. Hemrick-Luecke, J. Pharm. Pharmacol., **41**, No. 7, 493 (1989).
8. G. Yoshio, M. Ioshimi, A Nobutaca, and M. Kazuaki, Jpn. J. Pharmacol., **55**, No. 1, 93 (1991).
9. R. E. Heikkila, A. Hess, and R. C. Duvoisin, Science, **224**, 1451 (1984)
10. H. Kinemuchi, C. J. Fowler, and K. F. Tipton, Neurochem. Int., **11**, No. 4, 359 (1987).
11. J. W. Langston, P. Ballard, J. W. Tetrud, and I. Irwin, Science, **219**, 979 (1983).
12. G. H. Lowry, N. J. Rosebrough, A. L. Farr, and R. J. Randall, J. Biol. Chem., **193**, No. 1, 265 (1951).
13. C. J. Schmidt, E. Bruckwick, and W. Lovenberg, Eur. J. Pharmacol., **113**, No. 1, 149 (1985).
14. S. H. Snyder and J. T. Coyle, J. Pharmacol. Exp. Ther., **165**, No. 1, 78 (1969).
15. U. Trendelenburg, Naunyn-Schmiederbergs Arch. Pharmacol., **332**, 201 (1986),

EFFECT OF AMIRIDINE ON MPTP-INDUCED PARKINSON'S SYNDROME IN MONKEYS

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KEY WORDS: N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine; monkey; Parkinson's syndrome; amiridine

Antiparkinsonian drugs used in clinical practice are not sufficiently effective and give rise to many side effects. The search for new pharmacologic agents for the treatment of Parkinson's disease is thus extremely urgent and depends very much on the possibility of obtaining an adequate model of this pathological process. It has been found that the compound N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) gives rise to a symptom-complex typical of idiopathic parkinsonism both in man and in monkeys [7-9, 11, 12].

A new preparation for the treatment of nervous and mental diseases, amiridine (9-amino-2,3,5,6,7-hexahydro-1H-cyclopenta[b]quinoline monohydrate hydrochloride) has recently been developed at the All-Union Research Center for Safety of Biologically Active Substances [3-5]. The study of the mechanisms of action of this compound has shown that a shift of the region of activation of the K^+-channels of the excitable membrane in the direction of hyperpolarization. In vitro, this effect will evidently be equivalent to an increase in the membrane resting potential (RP), and it will consequently lead to more reliable generation and spread of the nervous impulse. Since RP of all cells, including those not electrically excitable, is maintained and regulated by potassium channels, amiridine can be expected to have a positive effect on impaired functions of nerve cells of all systems, whatever the factors causing their degenerative changes [9].

The aim of this investigation was to study the development of behavioral disturbances in monkeys receiving MPTP and to study the effect of amiridine on the symptoms thus produced.

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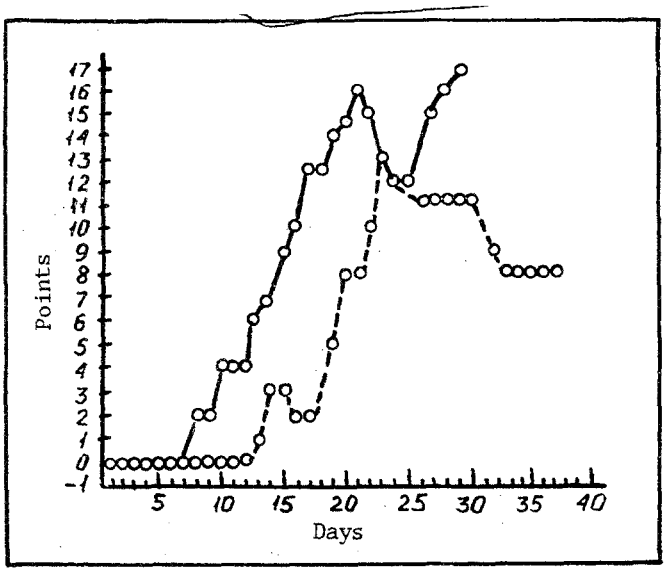

Fig 1. Changes in integral assessment of behavior of monkeys with a model of Parkinson's syndrome. Abscissa, days of experiment; ordinate, value of rating in points. Ratings for monkey Cheburashka indicated by continuous line, for Maika by a broken line.

EXPERIMENTAL METHOD

Experiments were carried out on two intact rhesus monkeys: Maika (a female aged 13 years and weighing 6 kg) and Cheburashka (a male aged 12 years weighing 8 kg). The animals were kept in separate cages under standard animal house conditions: temperature 27°C, 12 h of daylight and 12 h of darkness, water and food ad libitum.

MPTP hydrochloride ("Aldrich") was generously provided by Alabama State University (Birmingham). MPTP was injected intramuscularly in a dose of 0.2 mg/kg in 1 ml physiological saline, once a day on alternate days, between 3 and 6 p.m. Maika received 11 injections of MPTP (total dose 13.2 mg) in the course of 23 days, whereas Cheburashka received seven injections of MPTP (11.2 mg) in the course of 15 days. After cessation of the MPTP injections, on the appearance of marked symptoms of parkinsonism, the monkeys were given 250 mg L-dopa with water daily (Maika for 2 days starting from the 24th day of the experiment, Cheburashka for 3 days, starting with the 20th day of the experiment). Subsequent investigations were carried out in accordance with two different schedules: starting with the 24th day of the experiment Maika was given 1 ml of a solution of amiridine in a dose of 0.4 mg/kg intramuscularly, for 6 days, and then in a dose of 0.25 mg/kg for 8 days. After the end of amiridine administration, on the 38th day of the experiment, the monkey was sacrificed and autopsied. Cheburashka received 1 ml of physiological saline. This monkey died 14 days after the end of administration of MPTP, on the 29th day of the experiment.

Behavioral disturbances induced by injection of MPTP were assessed in points, using a clinical scale [10]. The following parameters of behavior were assessed: the level of motor activity (bradykinesia, akinesia), flexed and rigid postures, abnormal limb movements (dyskinesia), disturbance of balance, tremor, rigidity of the muscles, aphagia, adipsia, vocalization, stereotypy, and aggressiveness. The normal score was 0 points, the most severe symptoms of parkinsonism were rated at +16 points, maximal side effects during treatment at −8 points.

EXPERIMENTAL RESULTS

A graph showing changes in the integral assessment of the monkeys' behavior during the course of the experiment is shown in Fig. 1.

The latent period of the first manifestations of behavioral disturbances caused by injection of MPTP was found to differ in the two monkeys. For instance, the first manifestations of a change of behavior, with diminution of feeding activity, moderately severe general inertia, and bradykinesia (integral assessment + 3 points) in Maika were observed on the 14th day after the beginning of MPTP injections. By the 20th day of the experiment the severity of these disturbances had increased, the animal froze for a long time in the same posture, food consumption decreased, and aggressiveness disappeared almost completely (+8 points). On the 23rd day the monkey completely refused to eat, drank only from the experimenter's hands, the duration of the rigid postures increased to several minutes, and stereotyped horizontal movements of the head with a frequency of 0.1-0.2 Hz began to be observed (+13 points).

Injections of MPTP were then stopped and after the 24th day amiridine treatment began, accompanied during the first 2 days by L-dopa. On the 2nd day after beginning of treatment, worsening of behavior was not observed, the monkeys began to take small amounts of food, but the severe inertia and rigidity of the muscles still remained and limb movements began to be accompanied by tremor. The general state was rated at +11 points. Against the background of amiridine treatment the animal's state improved, the tremor decreased, and the motor and feeding behavior became more active. The animal's state stabilized at +6 points 10 days after the beginning of treatment, and no significant changes ensued before the end of the experiment.

The appearance of pathological symptoms caused by injection of MPTP in Cheburashka was distinguished by a shorter latent period and greater severity. By the 10th day, after four injections of the neurotoxin, this monkey showed a significant decrease in activity, bradykinesia, and depressed feeding activity (+4 points). On the 13th day these manifestations worsened to +6 points, the animal began to stoop, and the disturbances gradually increased. On the 16th day of the experiment, 1 day after the last injection of MPTP, and during the next 4 days the animal refused to eat, its general condition worsened sharply, the inertia increased, rigidity of the muscles, especially the limb muscles, was severe, and goal-directed movements were impossible. The animal froze for long times in a forced posture, lost its balance, and could not raise itself unaided. Movements were accompanied by a severe tremor, the animal responded only to strong stimuli, and the severity of the symptoms reached +14 points. Because of the animal's poor condition, the monkey was given L-dopa for 3 days. By the 23rd day of the experiment the animal's state showed some improvement, passive feeding behavior appeared, the aggressive reaction was strengthened, the monkey became more active, but the strong tremor still remained. On the 27th day the animal's condition suddenly worsened – rigidity gave way to paralysis of the limb muscles, and during attempts to move, a high-amplitude and low-frequency tremor was observed. The monkey could not raise itself, took food only from the experimenter's hands, and in very small amounts. Death occurred on the 29th day of the experiment.

Against the background of amiridine therapy in doses of 0.25-0.4 mg/kg gradual partial compensation of the behavioral disturbances took place after the end of the MPTP course. Although the state of the 2nd monkey (Cheburashka) at the time of the last injection of MPTP did not differ significantly from that of the other monkey, during the next 2 weeks the character of compensation of the disturbances differed greatly. The initial slight improvement in the animal's state can be explained as the result of L-dopa administration or by natural compensatory processes taking place in the brain. However, 1 week later, the condition of this monkey worsened sharply in the absence of amiridine treatment, and death occurred. Prolonged daily administration of amiridine to the 2nd monkey led to restoration of the various parameters up to 50-100% of the normal value, and this state lasted 1 week. Administration of amiridine thus causes gradual alleviation of the pathological disturbances induced by MPTP.

The mechanism of the compensatory action of amiridine on disturbances caused by injection of MPTP is not yet clear. This mechanism is evidently unconnected with any action on the dopaminergic system, for it has been shown that amiridine in vitro inhibits the synaptosomal uptake of DA only in high concentrations ($5 \cdot 10^{-5}$ M), and from this point of view it is much inferior to the powerful inhibitors of DA reuptake, nomifensine and mazindol [4]. Amiridine does not affect DA and MA levels in various structures of the rat brain and lowers the concentration of their metabolite dihydroxyphenylacetic acid [6]. Amiridine does not exhibit effects characteristic of stimulators of the dopaminergic system: it does not weaken the effects of reserpine, does not potentiate the action of dopamine agonists, and does not possess an anticataleptogenic action.

The mechanism of the positive effect of amiridine on motor functions in monkeys disturbed by administration of MPTP is thus more likely to be connected with an increase of the resting membrane potential, associated with its effect on potassium currents [9].

REFERENCES

1. Yu. V. Burov, T. N. Robakidze, L. V. Kadysheva and S. A. Suzanova, Byull. Éksp. Biol. Med., **111**, No 6, 612 (1991).
2. Yu. V. Burov, T. N. Robakidze, L. V. Kadysheva, et al., Byull. Éksp. Biol. Med., **111**, No. 6, 614 (1991).
3. Yu. V. Burov, Yu. N. Belyi, and T. N. Robikidze, Abstracts of Proceedings of the 1st Russian National Congress on Man and Medicine [in Russian], Moscow (1992), p. 548.
4. Yu. V. Burov, T. D. Baimanov, and N. I. Maisov, Byull. Éksp. Biol. Med., No. 4, 379 (1992).
5. M. Ya. Voloshin, Neirofiziologiya, **22**, No. 3, 401 (1990).
6. A. E. Voronin, T. N Robakidze, and I. I. Miroshnichenko, Current Trends in the Development of Biotechnology [in Russian], Moscow (1991), p. 48.
7. B. R. Bloem, I. Irwin, O. J. S. Buruma, et al., J. Neurol. Sci., **97**, No. 2-3, 273 (1990).
8. R. S. Burns, C. C. Chiueh, S. P. Markey, et al., Proc. Nat, Acad. Sci. USA, **80**, 4546 (1983).
9. Yu. Burov, Biol. Psychiat., **29**, 480S (1991).
10. C. E. Clarke, M. A. Sambrook, I. J. Mitchell, and A. R. Crossman, J. Neurol Sci., **78**, No. 3, 283 (1987).
11. G. C. Davis, A. C. Williams, S. P. Markey, et al., Psychiat. Res., **1**, No. 2, 249 (1979).
12. J. W. Tetrud and J. W. Langston, Acta Neurol. Scand., **126**, 35 (1989).

PROTECTIVE ROLE OF ESTRADIOL IN EXTREMAL STATES

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KEY WORDS: estradiol; hemorrhagic shock; cardiovascular system

Hormones and, in particular, corticosteroids and their synthetic analogs, in large doses, are widely used in extremal states [6, 18]. Information on their efficacy is contradictory in character. The possibility of using other types of steroid hormones, namely estrogens and androgens, in the treatment of extremal states has virtually not been investigated because of the absence of water-soluble forms suitable for intravenous injection.

Taking the above facts into consideration, it was decided to study the action of estradiol on survival and on some functional-metabolic parameters of the cardiovascular system of intact animals and of animals with hemorrhagic shock.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred rats of both sexes weighing 150-220 g and on dogs weighing 8-15 kg. Acute blood loss in the dogs and rats was induced by unrestricted bleeding from the femoral or carotid artery under pentobarbital anesthesia.

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