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MINUTE RHYTHMS OF HALOPERIDOL-INDUCED CATALEPSY IN RATS

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UDC 616.8-009.15-092.9"413"

KEY WORDS: haloperidol; catalepsy; chronopharmacology

Processes taking place in the brain have a precise temporal organization, and in turn, this affects fluctuations of activity of various neurotropic drugs. This state of affairs is confirmed by analysis of the as yet limited information in the literature on the role of the circadian rhythm of work of the brain in the action of psychotropic drugs [2]. During recent years shorter fluctuations, measured in minutes and tens of minutes, in the behavior and brain activity have assumed great functional importance [1, 3]. It is also important to take these fluctuations into consideration when the properties of psychotropic drugs and, in particular, the cataleptogenic effect of neuroleptics, are evaluated.

The aim of this investigation was to study rhythmic fluctuations of catalepsy after administration of haloperidol (HP).

EXPERIMENTAL METHOD

Experiments were carried out on 16 noninbred male albino rats weighing 200-300 g. For visual and graphic recording of the intensity of neuroleptic-induced catalepsy, we used our own modification of the holding onto a horizontal rod test [5]. The time during which the rat held on to the rod, hanging down from it and grasping the rod with its forelimbs, was counted. These determinations were carried out every minute for 2-4 h. HP was injected intraperitoneally in doses of 0.25 to 2 mg/kg. There were two series of experiments: In one of them the animals were given the same cataleptogenic dose of the neuroleptic in repeated tests. In the other series the effect of increasing doses of the drug was assessed after different times. In both series the interval between injections was not less than 4 days. Observations on each rat were repeated on average 5 times. In some investigations progressive accumulation of the drug took place in the course of one experimental day. The anticataleptogenic action of dopa (levodopa in a dose of 25, 50, or 100 mg/kg) was determined against the background of HP. The animals were kept in the animal house and allowed food and water *ad lib*. Natural illumination was provided and the experiments were carried out at the same time of day. There were altogether 70 experiments on two groups of rats: 42 experiments in winter (January-February) and 28 in spring (March-May).

The results of individual experiments were analyzed by the sliding means method [6]. To discover a periodic process the data were subjected to autocorrelation and spectral analysis on the Nairi-2 computer.

EXPERIMENTAL RESULTS

Under the influence of a cataleptogenic dose of HP (starting with a dose of 0.25 mg/kg)

Department of Pharmacology, Stavropol' Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR V. V. Zakusov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 101, No. 1, pp. 89-91, January, 1986. Original article submitted December 21, 1984.

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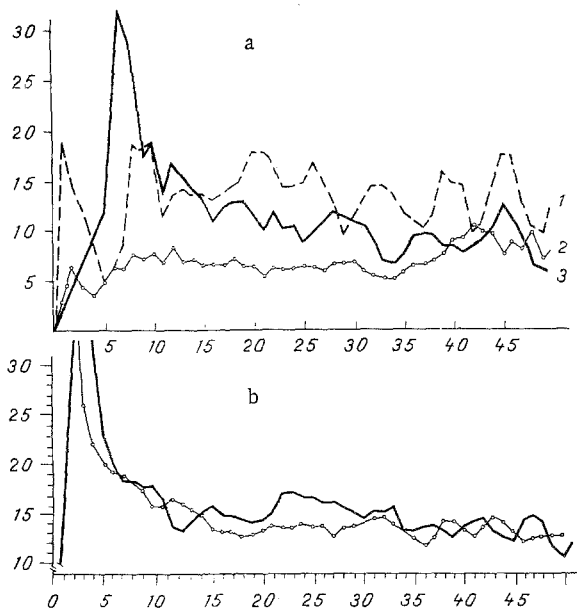

Fig. 1. Time course of catalepsy after injection of 0.5 mg/kg HP in different rats (a) and in repeated experiments on the same rat (b). Abscissa, time of recording (in min); ordinate, duration of catalepsy (in sec). a: 1, 2, 3) results of experiments on rats no. 15 (May 26, 1984), no. 12 (May 25, 1984), and no. 5 (February 14, 1984) respectively; b) data for rat no. 5 (March 19 and 29, 1984).

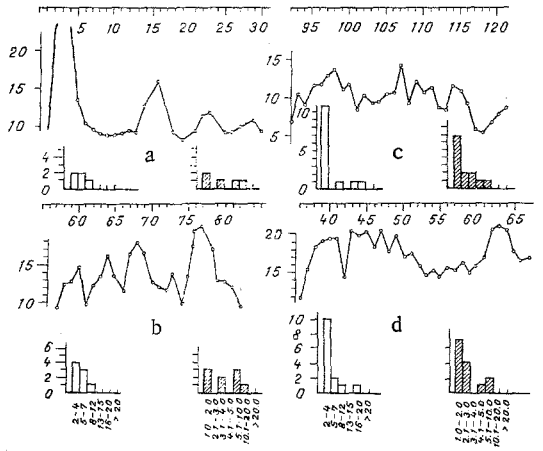

Fig. 2. Amplitude and temporal characteristics of different phases of haloperidol-induced catalepsy. a-c) Beginning, rise, and fall of catalepsy respectively in one experiment on rat no.14 after injection of 1 mg/kg of HP; d) disturbance of regular oscillations after injection of 25 mg/kg of levodopa at the height of catalepsy. Unshaded columns — histogram analysis of oscillating curve by period, shaded columns — the same, by amplitude. Values along axes of coordinates the same as in Fig. 1.

the rats acquired the ability of holding passively on to the horizontal rod with their forelimbs for several seconds. The total duration of catalepsy, estimated by means of this test,

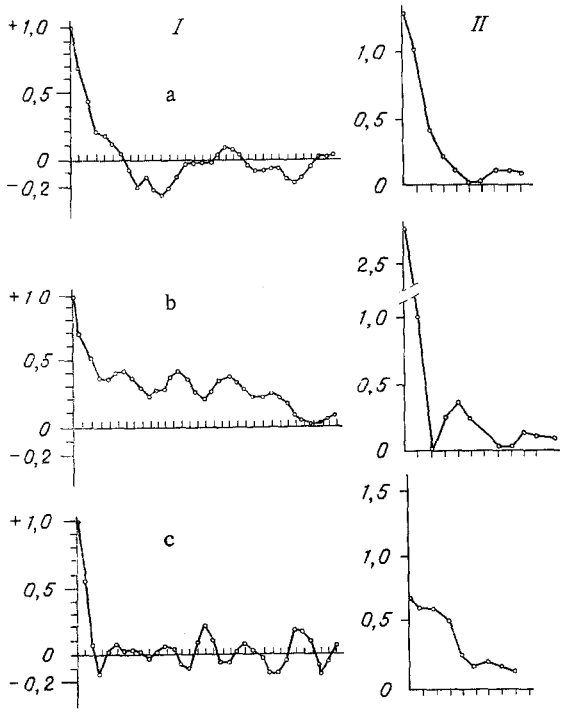

Fig. 3. Autocorrelation (I) and spectral (II) analysis of temporal structure of haloperidol-induced catalepsy after injection of increasing doses of haloperidol (rat no. 15): 0.25 mg/kg (a), 0.5 mg/kg (b), 1 mg/kg (c).

varied from 3 to 4 depending on several conditions: the dose of the drug, individual sensitivity and age of the animal, and season of the year. An increase in the dose of HP up to 1 mg/kg or more did not always prolong the catalepsy, and in some cases muscular relaxation increased, and in the late stages of recording the rats could not hold on to the rod. The optimal dose for study under the experimental conditions used was thus most frequently a dose of 0.5 mg/kg of HP.

Consecutive evaluation of catalepsy every minute demonstrated significant variation in its duration. On average (for 1 min), according to the results of the whole experiment, the duration of catalepsy was greater in the animals in spring (11.4±0.3 sec) and rather shorter in winter (7.9±0.2 sec). The results of neighboring determinations differed appreciably from one another. As a result, the graph showed a distinct oscillating curve (chronogram), which was then smoothed by the sliding mean method (Fig. 1).

The results of repeated determinations of the time course of haloperidol-induced catalepsy confirmed that it is a nonstationary oscillatory process. The primary chronogram, on visual inspection, had the appearance of an irregular sinusoid oscillation that differed in amplitude and period. Because of significant differences in the pattern of the chronograms, our first attempts to discover some general principle in their evolution were unsuccessful. In the first place, despite the use of a standard dose of HP, the shape of the curves differed appreciably for individual animals (Fig. 1a). Chronograms of repeated determinations of catalepsy in the same rat likewise did not coincide either in phase or in frequency (Fig. 1b). Such variability and the absence of an effective origin for counting prevented superposition of the chronogram or evaluation of the results of several experiments combined.

Analysis of the curves for separate experiments proved to be more informative. In conjunction with mathematical methods, this technique revealed certain principles governing the temporal organization of the pharmacological effect which, in our view, are important. In particular, it was observed that catalepsy passes through several stages, or phases, of development, and that the formation of marked catalepsy corresponds to typical reorganization of the oscillatory curve.

Analysis of the chronograms revealed the phasic structure of catalepsy, in which the

time of evolution of the pharmacological effect, the time of its maximal development and, finally, its decay could be determined. The first phase was characterized by extreme instability of the process, in the form of an irregular rhythm, sometimes with high-amplitude oscillation. It will be noted that in most animals (58.3%) haloperidol-induced catalepsy began with a dramatic peak-like surge of the oscillatory curve. Paradoxically as it may seem, in this case the duration of catalepsy was longest (from 16.2 to 43.2 sec) in the first 2-10 min after injection of HP (Figs. 1b and 2a), and this was followed by a series of oscillations which differed considerably in both period and amplitude. According to the results of histogram analysis, this phase was dominated by oscillations with periods of 2-4, 5-7, and 8-12 min (Fig. 2a).

The initial phase of catalepsy, according to our estimates, ended by the time of formation of a rhythm with a well-organized frequency and amplitude. The time of onset of the second phase varied appreciably in different animals, and irrespective of dose it could last up to 50-60 min.

Distinct synchronization of the rhythm, in the form of stabilization of the pattern of the chronograms, and regularity of the period and amplitude of the waves, corresponded to development of the maximal pharmacological response. In the phase of stabilization of the effect the number of waves within the band from 2-4 to 5-7 min increased (Fig. 2b). At the same time, the waves in the group of experiments conducted in spring were more pronounced than those in winter.

The final phase of the process, namely extinction of catalepsy, was characterized by increasing breakdown of the oscillatory process. The pattern of the chronograms begans to be dominated by fast waves of low amplitude (Fig. 2c). Not only natural weakening of catalepsy, but also its destruction by dopa, injected at the climax of the response to HP, followed a similar type of course (Fig. 2d).

By the use of increasing doses of HP it was possible to determine which phenomena accompany the development of the most severe catalepsy: the higher the dose, the clearer the increase in the number of waves with a period of 5-7 min and of sufficient amplitude. This rule, and the actual period of the oscillations in haloperidol-induced catalepsy were confirmed by the results of autocorrelation and spectral analysis (Fig. 3).

Consequently, the study both of phasic features in the course of the pharmacological effect and of its changes under the influence of increasing doses of the neuroleptic revealed the same tendency: The formation of marked catalepsy is a feature of the appearance of regular fluctuations of muscle tone.

The rhythmic fluctuations of haloperidol-induced catalepsy, which we established, may be determined by fluctuations of muscle tone that are natural [7, 8], but emphasized by the drug. The participation of certain pharmacokinetic factors in this process, such as temporal oscillations of activity of the microsomal enzymes of the liver [4], likewise cannot be ruled out. Irrespective of the nature of the phenomenon the results indicate the undoubted informativeness of evaluation of the time course of the neuroleptic effect.

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