ELECTROENCEPHALOGRAPHIC INVESTIGATION OF THE ANTAGONISM BETWEEN 5-HYDROXYTRYPTAMINE AND CHLORPROMAZINE AND TRIFTAZIN (Trifluoperazine)

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The study of the role of serotonin in the functional activity of the central nervous system based on the direct administration of serotonin is difficult because of the low permeability of the blood-brain barrier to this amine [18]. More constant effects have been observed as a result of the administration of the serotonin precursor 5-hydroxytryptophan which, when it enters the brain, undergoes decarboxylation there, giving rise to an increase in the serotonin content in the brain [11, 14].

As V. V. Zakusov has emphasized [1], an important aspect of the study of the biological role of serotonin is the investigation of its antagonists—substances of varied chemical structure preventing the manifestation of the physiological effects of serotonin. Phenothiazine compounds [4, 12] have been found to possess marked antiserotonin activity, and some investigators [7, 19] associate the tranquilizing effect of the phenothiazine derivatives with this activity. However, most of the findings obtained during the study of this problem have been gathered from experiments on isolated organs [13], i.e., experiments conducted in technical conditions inadequate for the solution of this problem. For these reasons it is essential to study the effects of the tranquilizers of the phenothiazine series on the central action of 5-hydroxytryptamine (5-HT).

The present paper gives the results of experiments carried out to study the influence of 5-HT on the electroencephalogram (EEG) and the effects of chlorpromazine and triftazin* in relation to the electroencephalographic phenomena of 5-HT.

EXPERIMENTAL METHOD

Chronic experiments were conducted on 40 noncurarized, unanesthetized rabbits. Phonograph needles inserted into the bone or bent silver electrodes introduced epidurally were used as cortical electrodes. For insertion into the subcortical regions (hippocampus, reticular and antero-ventral nucleus of the thalamus, reticular formation of the mesencephalon [16]), electrodes made of insulated nichrome wire, 100-125 μ in diameter, were used, and the distance between the electrodes was 1 mm. Their position was verified after death [12]. The EEG and ECG (lead 2) were recorded on an 8-channel ink-recording electroencephalograph (Alvar). The 5-HT was injected intravenously at constant rate (1.67 mg/kg/min).

* Triftazin is a compound identical with Stelazine; it is the dihydrochloride of 10- 3-(1-methylpiperazinyl-4)-propyl-2-trifluoromethylphenothiazine. The compound was obtained under S. V. Zhuravlev's direction at the Institute of Pharmacology and Chemotherapy, USSR Academy of Medical Sciences, and its pharmacological properties were studied by K. S. Raevskii, B. I. Lyubimov, and T. A. Klygul' [2].

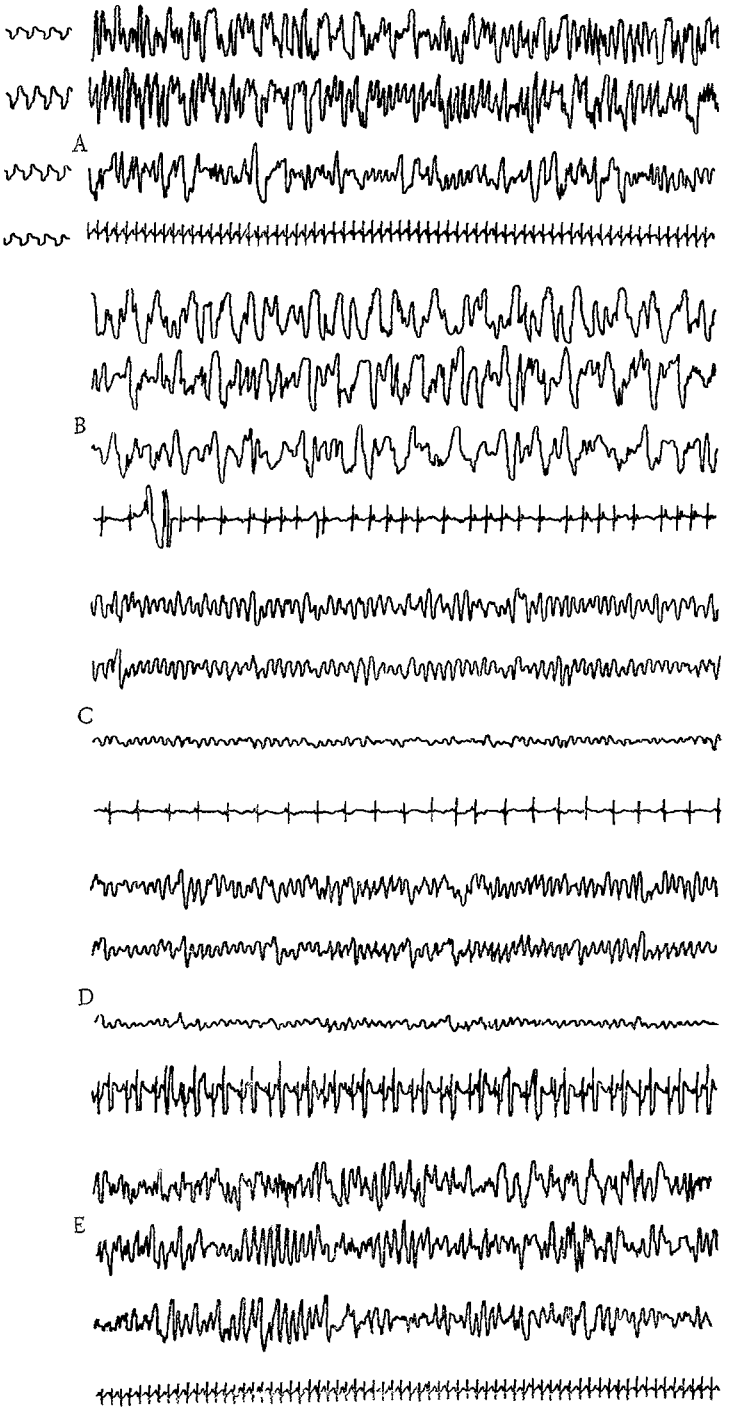

Fig. 1. Effect of 5-hydroxytryptophan on the EEG and ECG. From top to bottom: EEG of right optic and associative and left anterior sensorimotor areas of the rabbit's cerebral cortex; ECG. A) Before administration of preparations; B) 12th min; C) 23rd min; D) 40th min of infusion; E) 3 h after infusion. Calibration 50 μV.

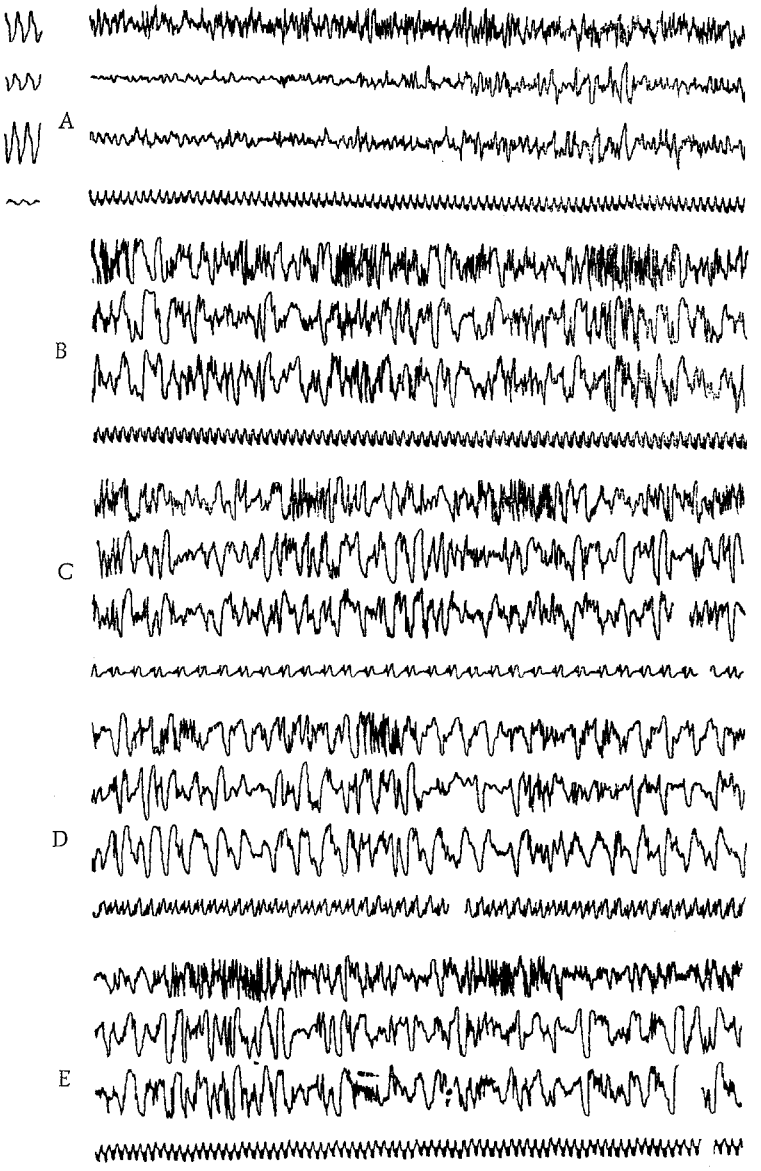

Fig. 2. Blocking effect of chlorpromazine on activation of the EEG caused by 5-hydroxytryptophan. Leads the same as in Fig. 1. A) Before administration of preparations; B) 5 min after administration of chlorpromazine in a dose of 5 mg/kg; infusion started; C) 25th min; D) 55th min of infusion; E) after end of infusion of 5-hydroxytryptophan in a dose of 120 mg/kg. Calibration 100 μV.

EXPERIMENTAL RESULTS AND DISCUSSION

In all the experiments 5-HT produced lasting and constantly reproducible changes in the EEG (Fig. 1). Between 20 and 25 min (in some experiments between 18 and 28 min) after the beginning of the infusion the amplitude of the cortical biopotentials fell to 15-25 μV; in the anterior portions of the cortex high-frequency waves appeared —up to 20 cps (often against the background of regular low-amplitude waves with a frequency of 4-5 cps), and in the posterior portions potentials of regular shape and a frequency of 4-5 cps were seen. Besides the considerable decrease in the amplitude, the pattern of activation of the EEG described above differed from the activation reaction characteristic of the action of cholinomimetic drugs by the preservation of individual slow waves of high amplitude. Activation of the EEG took place after 22.25±1.61 min (from the beginning of infusion), corresponding to a dose of 37.15±2.68 mg/kg. The duration of this phase varied from 1.5 to 2.5 h. Its appearance during the action of 5-HT was sometimes preceded by a phase of slow (up to 2-3 per sec), high-amplitude (up to 200-250 μV) potentials

(Fig. 1B), usually beginning after the 9th-12th min and continuing until the onset of the second phase (of the activation reaction). However, the first phase, unlike the second, was not clearly defined in all the animals.

In the experiments on the rabbits with the implanted electrodes, together with the disappearance of the slow, high-amplitude potentials in the cortex, the mesencephalic reticular formation, and the nonspecific nuclei of the thalamus during the second phase, pointed potentials with a frequency of up to 20-30 cps appeared in the hippocampus. These discharges were evidently analogous to the hippocampal spikes described earlier [9].

The changes in the EEG listed above were accompanied by marked changes in the animals' general behavior: between the 20th and 30th min of infusion the rabbits became restless, they began to breathe rapidly, and tugged at their straps; they developed a tremor and convulsive spasms of individual muscles of the limbs and trunk, and made stereotyped movements of the head.

In the next series of experiments the effect of triftazin and chlorpromazine on the action of 5-HT described above was studied. Since preliminary tests showed that the dose of triftazin causing prolonged (up to 2.0-2.5 h), slowing of the background activity and an increase in the amplitude of the dominant rhythm, with the appearance of spindles, was 5 mg/kg (intravenously), the rabbits were given 5 mg/kg of triftazin 10 min before the infusion of 5-HT began. The experiments showed that 5-HT, when given against this background, did not cause activation at the times established previously; despite the continued infusion, activation developed only after 45.2±2.68 min ($P < 0.001$). These changes in the times of onset of activation of the EEG corresponded to an increase in the activating dose of 5-HT from 37.15±2.68 to 75.48±4.36 mg/kg.

Injection of chlorpromazine in a dose of 5 mg/kg 10 min before the beginning of infusion completely prevented the activating effect of 5-HT. Despite the continued infusion of 5-HT, the high-amplitude slow waves remained dominant in the pattern of the background bioelectrical activity even after infusion of 120 mg/kg, i.e., of a dose more than 3.2 times larger than the usually effective dose of 5-HT (Fig. 2)*. If chlorpromazine was injected in a dose of 3 mg/kg, it did not cause the complete blocking of the effect of 5-HT, but modified the times of its onset: signs of activation, as against the background of administration of triftazin (5 mg/kg), usually appeared after 50 min, but the reaction was even less marked than against the background of triftazin.

Similar patterns were seen when the animals' general behavior was observed. When 5-HT was given against the background of chlorpromazine, it did not cause the characteristic picture of excitation at the 20th-30th min of infusion. Not until the 50th-60th min did the rabbit give occasional convulsive spasms of the head, although its behavior was dominated by signs of depression characteristic of the effect of chlorpromazine. Against the background of triftazin these spasms were rather stronger, although they were much weaker than in the control animals.

The antagonistic action of the phenothiazine derivatives in relation to the effects of 5-HT was prevented by the preliminary administration of iproniazid. If iproniazid was injected intravenously in a dose of 50 mg/kg 18 h before administration of chlorpromazine or triftazin, the action of 5-HT was considerably strengthened: at the 30th-40th min of infusion the rabbit developed marked excitation or even actual fits. The moment of onset of fast, low-amplitude waves on the EEG coincided with the appearance of the initial signs of excitation, although as these continued to develop it became increasingly difficult to record the EEG.

Analysis of the effect of these tranquilizers on the EEG showed that they did not prevent the bradycardia regularly produced in rabbits by 5-HT; they merely reduced its intensity very slightly.

Conflicting opinions have been expressed in the literature on the effect of 5-HT on the bioelectrical activity of the brain. Some authors [5], for example, emphasize the ability of 5-HT to cause the appearance of high-voltage, slow waves with the disappearance of the fast cortical rhythms, whereas others [16], on the contrary, report only fast, low-amplitude rhythms after the administration of this substance; finally, some investigators observed both effects after aministration of 5-HT; both synchronizing and desynchronizing [9, 14]. It is difficult to compare the results obtained by different authors, for they were obtained after the administration of different doses of the particular drug by different ways to different animals. Another essential cause of the differences between these results was that 5-HT causes changes in the hemodynamics, and the severity of these changes depends on the mode of administration of the drug. In experiments preceding the study of the electroencephalographic effects of 5-HT, the author showed that when it was given in a single dose of 50 mg/kg intravenously, the arterial pressure fell to 50-60 mm. Since a

* When higher doses of 5-HT were given the animals usually died.

rapid fall in the arterial pressure to this level and is known to lead to a disturbance of the blood supply to the brain [10] and, in consequence of this, to the appearance of secondary changes in the EEG in the form of slow, high-amplitude potentials, the 5-HT was injected as a single dose immediately, but by the method of slow intravenous infusion. In these circumstances the arterial pressure was stable (maximal decrease 10 mm).

When injected in this manner, 5-HT caused characteristic changes in the EEG, and these may be represented schematically in two phases: the first as regards the time of its appearance (9th-10th min of infusion)—the phase of high-amplitude, slow potentials, which was clearly defined not in all cases, and the second—the phase of fast, low-amplitude oscillations, characterized by stability of its times of appearance, its degree, and its duration; it was accompanied by characteristic changes in the animals' behavior. In its general features the picture observed was similar to the changes in the EEG described by Domer and Longo [9] after administration of 5-HT, although these workers did not emphasize the remarkable stability of the times of appearance of the phase of low-amplitude potentials. This was probably because in all their experiments the speed of infusion was the same regardless of the weight of the animals.

Bearing in mind the foregoing remarks on the activating effect of 5-HT, and also because of the fact that the times of its appearance were close to the times of maximal increase in the serotonin content of the brain following administration of 5-HT [6], it was decided to use the activating effect of this substance as one of the tests for studying the central antiserotonin effect of the investigated phenothiazine derivatives—chlorpromazine and triftazin. It was found that both compounds had a marked antagonistic action on this activation reaction; this was revealed by the increase in the dose required to obtain it (5 mg/kg against the background of triftazin), or even by the complete blocking of activation (when the same dose of chlorpromazine was given). The results indicating that the antiserotonin effect of chlorpromazine is stronger than that of triftazin are in agreement with the results obtained when the influence of these drugs was studied on the behavioral changes produced in rats and mice by 5-HT [4, 17].

The physiological mechanism of the antagonistic relationships thus discovered require further study. Experiments involving section of the brain stem at various levels showed that the electroencephalographic effect of 5-HT is connected with its influence on the mesencephalon [16]. On the other hand, the mesencephalic reticular formation is known to play an important role in the mechanism of action of the phenothiazine derivatives [8, etc.]. It may evidently be postulated that the antagonistic influence of chlorpromazine and triftazin on the electroencephalographic effects of 5-HT is brought to bear at the level of the serotoninergic structures of the mesencephalic reticular formation and that this component plays a decisive role in the mechanism of action of the phenothiazine derivatives in addition to the blocking of the adrenergic structures of the reticular formation described previously.

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