Effects of Azaleptine and Its New Derivative Seleptine on Spontaneous EEG Activity and the Activation Reaction in Rabbits

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 119, № 3, pp. 252-255, March, 1995
Original article submitted April 26, 1994

The neuroleptic azaleptine and its new derivative seleptine with anticonvulsive properties were examined in a chronic experiment on rabbits for their comparative effects on the spontaneous electroencephalogram (EEG) and the activation reaction in the sensorimotor cortex and dorsal hippocampus. Azaleptine induced synchronization of electroencephalographic activity in all frequency ranges of both cortical and hippocampal EEGs, while seleptine induced desynchronization in the cortical EEG and synchronization in the δ, θ, and β ranges of the hippocampal EEG. Both compounds prevented the activation or altered its pattern, leading to a decrease in the power of the β range rather than increasing it as is normally observed.

Key Words: *azaleptine; seleptine; neuroleptics; anticonvulsants; total electroencephalogram; activation reaction*

Azaleptine (8-chloro-11-(4-methyl-1-piperazinyl)-5H-dibenzo-[b,e][1,4]-diazepine), which has been successfully used in medical practice, is a typical neuroleptic with a unique neuropharmacological profile [4,8]. Its new structural analog seleptine (8-chloro-11-(4-methyl-1-piperazinyl)-5-acetylamino-dibenzo-[b,e][1,4]-diazepine) is regarded as a potential anticonvulsant with possible psychotropic properties.

In this study on rabbits, we compared the effects of the two compounds on spontaneous electric activity of the sensorimotor cortex and dorsal hippocampus as a test for alterations in the general excitability of the brain and in the functional status of neurotransmitter systems.

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MATERIALS AND METHODS

This chronic experiment was carried out on rabbits (body weight 2.3-3.5 kg), 6 animals in each group, implanted stereotaxically with bipolar nichrome electrodes under Nembutal anesthesia (30 mg/kg after Aminazine [chlorpromazine] administration). The recording electrodes were implanted into the sensorimotor area of the neocortex (AP 2-3; L 2; H 2) and into area CA3 of the dorsal hippocampus (AP 5.5-7; L 5.2-6.5; H 3.5), the stimulating electrodes were implanted into the mesencephalic part of the reticular formation (nucleus cuneiformis, pars dorsalis; AP 13.5; L 2.4; H 7), while the indifferent electrodes were placed in the nasal bone and over the visual cortex in the occipital bone. Tests were started 5-7 days postimplantation.

The total electroencephalogram (EEG) was recorded bipolarly in nonrigidly fixed animals in a shielded soundproof darkened chamber. The EEG

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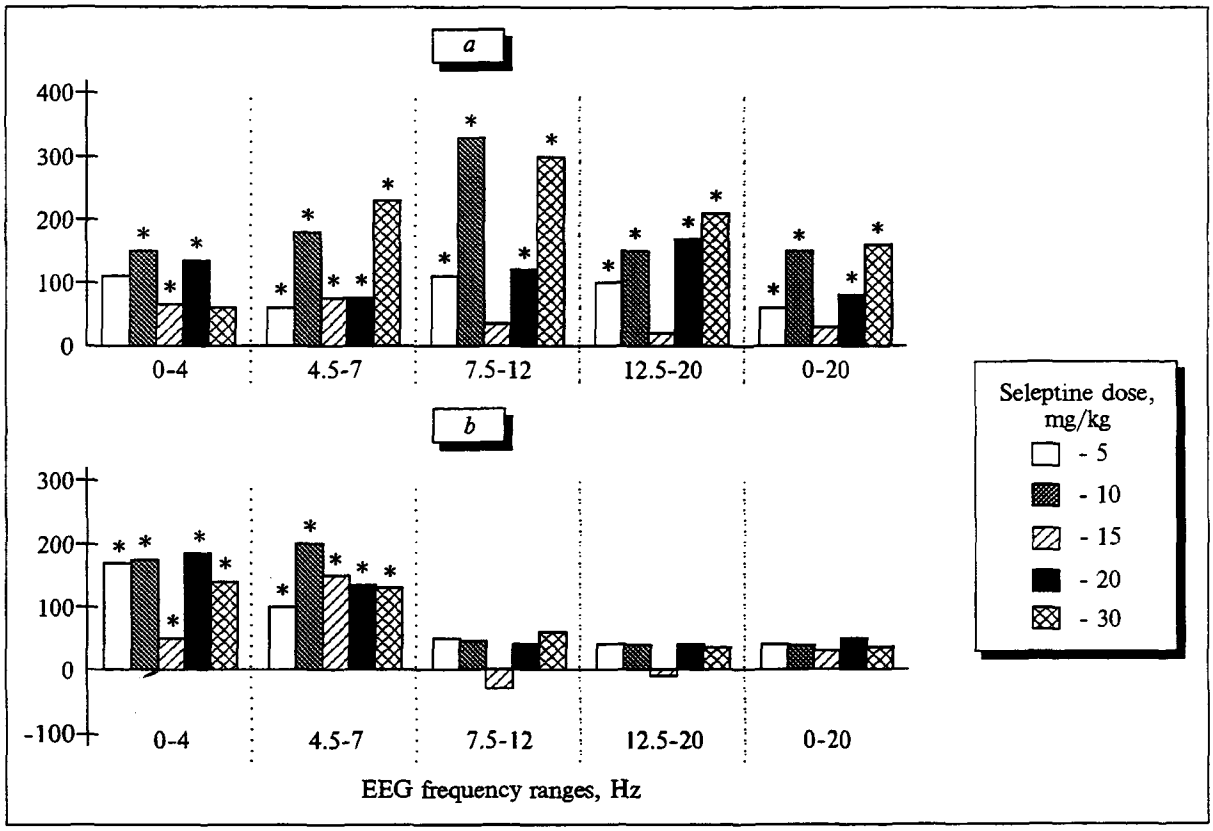

Fig. 1. Effects of azaleptine in various doses on the power of the total EEG and of individual frequency ranges 60 min postinjection. *a*) sensorimotor cortex; *b*) dorsal hippocampus. Ordinate: percentage changes in the power of EEG ranges over background values. *$p<0.05$ (ANOVA test).

signal was monitored on the electroencephalograph and concurrently on a graphic display. The power spectrum of the EEG was evaluated automatically using a KAMAK system and an Elektronika-80 computer (both Russian-made), with an analytical epoch of 2 sec and a quantization period of 8 msec. EEG power spectra were calculated from 256 dots using a fast Fourier transform algorithm. Based on our own data and those reported in the literature [2,7,12], the following stimulation parameters were selected as optimal for eliciting an activation reaction: flashes of directional light (from an arc photoflash), auditory stimulation with a tone of 300 Hz for 2 sec, and electric stimulation of the reticular formation (RF) with square pulses of 1 msec delivered at a frequency of 100 Hz for 10 sec. Stimulation intensities were chosen individually for each animal, and their working values used in the tests were equal to 110-115% of those just sufficient to elicit a well-defined activation reaction.

The effects of azaleptine and seleptine (both synthesized at the Novokuznetsk Chemical-Pharmaceutical Institute) on the activation reaction were evaluated according to the following scheme: 1) background patterns and the power spectrum of the EEG were recorded as well as threshold values of sensory and electric stimulations for activation reaction induction; 2) dissolved azaleptine or seleptine (200 mg + Tween 80 + 5 ml of physiological saline) was injected intraperitoneally in a dose of 5, 10, 15, 20, or 30 mg/kg; 3) the presence or absence of an activation reaction was checked 30 and 60 min later at the working values of stimulation intensity. If there was no reaction, the dose used was considered to be effective in preventing the activation reaction, and further tests were then run 90 and 120 min postinjection.

The data were processed in a conventional manner, by analyzing both the total power of the EEG spectrum (0-20 Hz) and the power of individual ranges [6]: δ (0-4 Hz), θ (4.5-7 Hz); a (7.5-12 Hz), and β (12.5-20 Hz). For analysis, percentage changes in the EEG power spectra caused by different azaleptine and seleptine doses relative to their background values were considered. The results were treated statistically by Student's *t* test and ANOVA software.

RESULTS

Before dosing, all rabbits were in a calm wakeful state with the eyes open. The EEGs recorded from

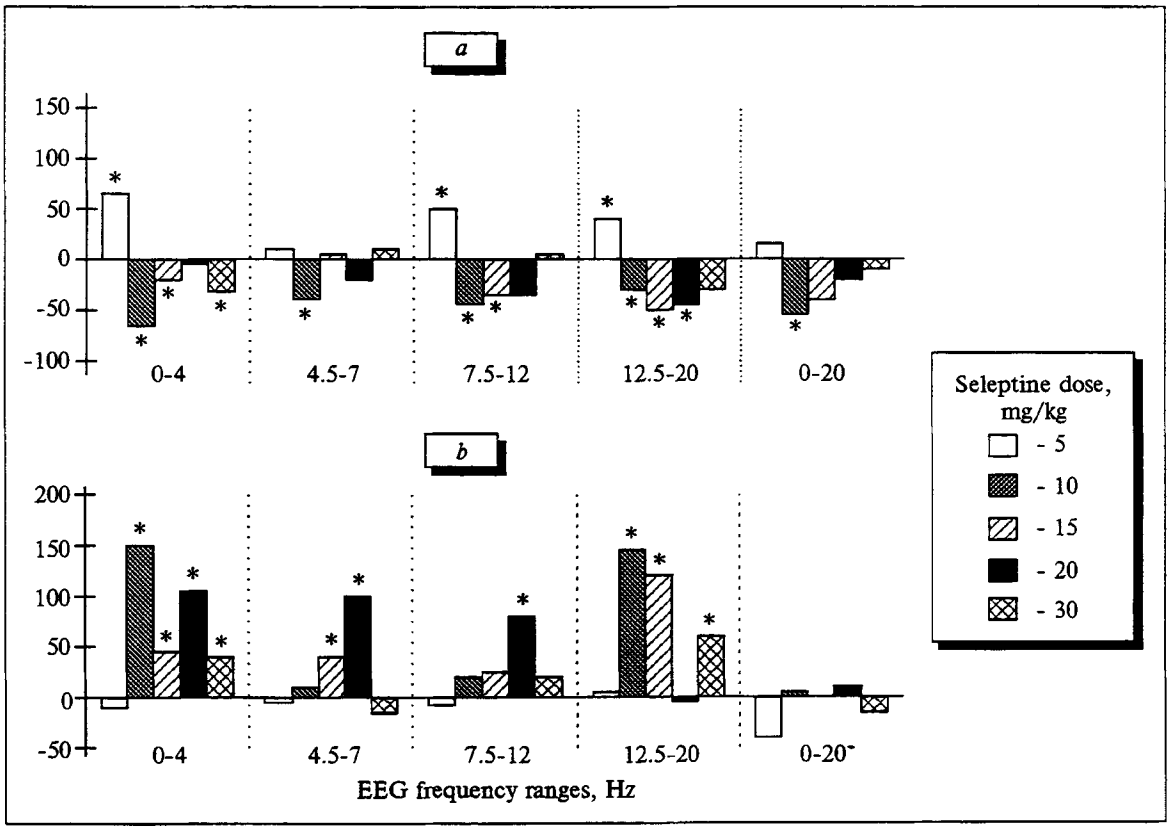

Fig. 2. Effects of seleptine in various doses on the power of the total EEG and of individual frequency ranges 60 min postinjection. Same designations as in Fig. 1.

the sensorimotor area of the neocortex were characterized by irregular activity of low amplitude (20-25 μV), with an obvious predominance of slow δ and θ waves in the EEG power spectrum. The dorsal hippocampal EEGs contained, along with the θ rhythm of 40-60 μV and a high-frequency β activity, some solitary high-amplitude (up to 100 μV) δ waves or whole areas of the δ rhythm up to 10 sec in duration.

The intraperitoneally administered azaleptine exerted an appreciable effect on the EEG pattern. Consistent findings for the azaleptine-treated rabbits may be described as follows.

Sensorimotor area of the neocortex: increased powers of the δ, θ, and α ranges, observed as early as 30 min after azaleptine injection at 10-30 mg/kg, followed by further increases in the power of these ranges in the next 30 min together with an increase in the power of the β range and, as a consequence, in that of the total EEG at min 60 postinjection (Fig. 1, *a*), with a return to the initial pattern of the EEG power spectrum by minute 120 postinjection.

CA3 area of the dorsal hippocampus: increased powers of the δ and θ ranges, even after the lowest dose (Fig. 1, *b*), followed by a return to the initial pattern of the EEG power spectrum by minute 90 postinjection.

Seleptine, unlike azaleptine, increased the power of the δ range in the sensorimotor cortex only in the lowest dose (5 mg/kg). In higher doses (10-30 mg/kg) it reduced the total EEG power in the cortex, the largest contribution to the reduction being made by decreases in the powers of the δ, α, and β ranges (Fig. 2, *a*). In the dorsal hippocampus, no significant changes in the total EEG power were detected, although the powers of the θ, β, and (more frequently and to a greater extent) δ regions were increased by doses of 10-30 mg/kg (Fig. 2, *b*).

These tests indicate that both azaleptine and seleptine alter the EEG pattern during the activation reaction to presented stimuli of various modalities and can even suppress the reaction completely. ED_{50} values, calculated as described by Kam Pui Fung [9], were 11.7±1.7 and 14.2±3.8 mg/kg for azaleptine and 10.2±2.2 and 10.8±3.0 mg/kg for seleptine with sensory stimulation and electrostimulation of the RF, respectively.

Statistical treatment of the EEG power spectra recorded during the activation reaction permitted detection of the reversal, brought about by both

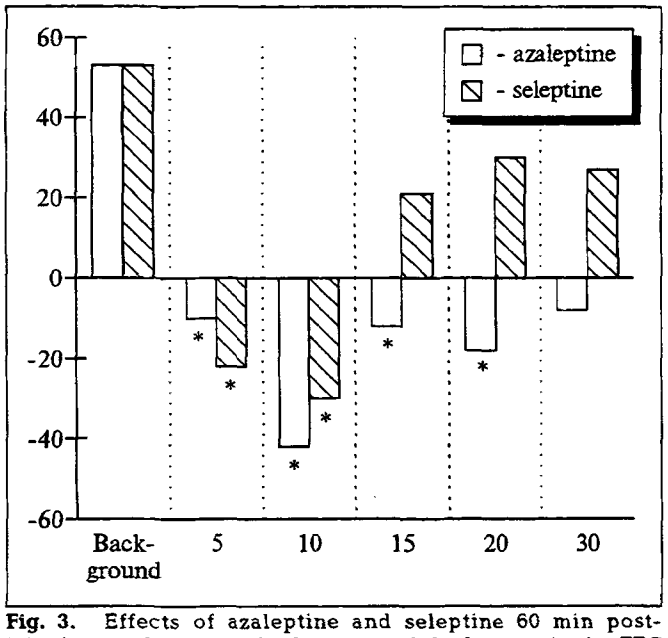

Fig. 3. Effects of azaleptine and seleptine 60 min post-injection on the change in the power of the b range in the EEG of the sensorimotor cortex recorded during the activation reaction to an auditory stimulus. Ordinate: percentage change in the power of the β range over its value immediately before the activation reaction. *$p<0.05$ (Student's t test).

compounds, of shifts in the power of the β range in the neocortical sensorimotor area in response to sensory stimuli: as illustrated in Fig. 3, a dose-dependent decrease in the power of this range occurred rather than the increase which is typical of background EEG activity and which specifically reflects the activation of the neocortex by the RF [3,12]. A similar tendency was observed for both the pattern of sensorimotor cortex activation in response to electrostimulation of the RF and the pattern of the hippocampal response to stimuli of any modality.

Analysis of the central effects produced by azaleptine confirmed the previously reported marked neuroleptic properties of this compound [6,10]; thus, azaleptine synchronized the sensorimotor neocortical and dorsal hippocampal EEGs in the lower frequency ranges (thereby increasing the total EEG power) while interfering with the activation reaction to presented stimuli of various modalities. These effects of azaleptine are presumed to stem from its cholinolytic and adrenolytic activities, leading to a relatively enhanced efficiency of the brain serotoninergic system [11], and from its high affinity for the serotonin receptors 5-HT_{1A} and 5-HT_2, because of which it can stimulate this neurotransmitter system directly [8].

The impact of seleptine on the functional status of the brain is less straightforward: as the results presented above indicate, this compound induced EEG desynchronization in the sensorimotor cortex, which is a characteristic feature of high-level wakefulness [3], while suppressing the activation reaction; concurrently, EEG synchronization in the lower frequency ranges developed in the hippocampus, associated with an inhibited state of the latter [2,3]. In addition to anticonvulsive activity, seleptine, in contrast to azaleptine, is likely to possess properties of tranquilizers that lower the excitability of subcortical structures responsible for emotional reactions (primarily of the limbic system) and interfere with the interaction of these structures with the neocortex [4] without affecting the activity of the cerebral cortex itself. Such actions of seleptine may be due to its adrenolytic activity [5,7] as well as to its ability to enhance inhibitory processes mediated by the GABAergic neurotransmitter system.

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