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Pharmacology, Biochemistry and Behavior 76 (2003) 169–178
PHARMACOLOGY BIOCHEMISTRY AND BEHAVIOR
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Interaction of astemizole, an H_1 receptor antagonist, with conventional antiepileptic drugs in mice

Mariusz Świąder[a], Marian Wielosz[a], Stanisław J. Czuczwar[b,c,*]

[a]*Department of Pharmacology and Toxicology, Medical University, 20-090 Lublin, Jaczewskiego 8, Poland*
[b]*Department of Pathophysiology, Medical University, 20-090 Lublin, Jaczewskiego 8, Poland*
[c]*Isotope Laboratory, Institute of Agricultural Medicine, 20-950 Lublin, Jaczewskiego 2, Poland*

Received 14 April 2003; received in revised form 19 July 2003; accepted 21 July 2003

Abstract

Histamine is one of the aminergic neurotransmitters, playing an important role in the regulation of a number of physiological processes. There are several subtypes of histamine receptors—H_1, H_2, H_3 and the recently discovered H_4. H_1 receptors exist on mast cells, basophils, enterochromaffin cells and in the central nervous system, being located postsynaptically. H_1 receptor antagonists, including classical antiallergy drugs, occasionally have been expected to induce convulsions in children and epileptics. The aim of this study was to evaluate the effects of astemizole-given intraperitoneally, singly or for 7 days on the anticonvulsant activity of antiepileptic drugs (AEDs) against maximal electroshock (MES)-induced convulsions in mice. The following AEDs were administered intraperitoneally: valproate magnesium, carbamazepine, diphenylhydantoin and phenobarbital. Adverse effects were evaluated in the chimney test (motor performance) and passive avoidance task (long-term memory). Brain and plasma levels of AEDs were measured by immunofluorescence. Astemizole (a single dose and following a 7-day treatment at 2–6 mg/kg) reduced the threshold for electroconvulsions, being without effect upon this parameter at lower doses. Astemizole (1 mg/kg) did not significantly alter the protective effect of AEDs against MES (after acute and 7-day administration). Also, acute astemizole (2 mg/kg) remained ineffective in this respect. Astemizole (2 mg/kg), following chronic administration, significantly reduced the protective efficacy of phenobarbital and diphenylhydantoin, reflected by an increase in their ED_{50} values (50% effective dose necessary to protect 50% of animals tested against MES) from 21.1 to 34.0 mg/kg and from 10.4 to 19.2 mg/kg, respectively. Astemizole (2 mg/kg) did not alter the protective activity of the remaining AEDs. Moreover, astemizole (2 mg/kg) did not influence the free plasma levels and brain concentration of the studied AEDs. Also, this H_1 receptor antagonist did not impair long-term memory or motor coordination when given acutely. However, 7-day treatment with astemizole (2 mg/kg) significantly decreased TD_{50} (50% toxic dose required to induce motor impairment in 50% of animals) value of phenobarbital, being without effect on carbamazepine, valproate and diphenylhydantoin in this respect. Similarly, phenobarbital and diphenylhydantoin, administered alone at their ED_{50}s against MES, or combined with astemizole, disturbed long-term memory in mice. The results of this study indicate that astemizole may need to be used with caution in epileptic patients.

Keywords: Astemizole; Antiepileptic drugs; Electroshock maximal; Drug interactions; Seizures

1. Introduction

Histamine is a neurotransmitter in the mammalian central nervous system (CNS) (Schwartz et al., 1991; Wada et al., 1991). There are several types of histamine receptors in the brain: H_1, H_2, H_3 and the recently described H_4. One CNS role of histamine in the brain has been suspected to inhibit seizures through H_1 receptors, but the roles of central histamine receptors in convulsions are still uncertain.

These suggestions were supported by experimental studies and clinical case reports. Gerald and Richter (1976) observed various effects of histaminergic agents on the susceptibility of mice to pentylenetetrazol-induced minimal (clonic) and maximal (tonic) seizures. Moreover, Tuomisto and Tacke (1986) indicated that central histamine might be important in the inhibition of maximal electroshock seizures (MES) in rats. Onodera et al. (1992) reported that brain histamine levels in epilepsy-prone rats were significantly lower than in epilepsy-resistant ones. Furthermore, L-histidine and metoprine increased pentyl-

* Corresponding author. Department of Pathophysiology, Medical University, 20-090 Lublin, Jaczewskiego 8, Poland. Tel.: +48-81-7425835; fax: +48-81-7425828.
E-mail address: czuczwar@galen.imw.lublin.pl (S.J. Czuczwar).

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doi:10.1016/S0091-3057(03)00212-0

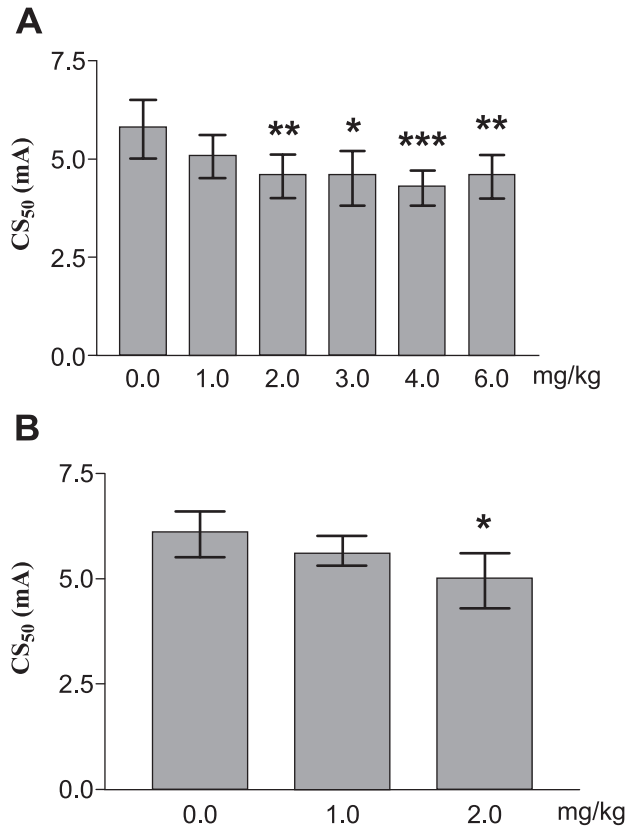


Fig. 1. Effects of acute and chronic administration of astemizole upon the electroconvulsive threshold in mice. (A) Astemizole in a single dose was given intraperitoneally 30 min before testing. (B) Astemizole after chronic administration (once daily intraperitoneally for 6 days), was also given on the seventh day, 30 min before testing. The control group received intraperitoneally 1% Tween 80. To estimate the convulsive threshold, at least four groups of mice (eight animals per group) were challenged with electroshocks of various intensities. The data are CS_{50} values (current strength of 50% with 95% confidence limits), calculated and compared according to Litchfield and Wilcoxon (1949). *P<.05, **P<.01, ***P<.001.

enetetrazol seizure threshold in mice (Scherkl et al., 1991b), inhibited seizure duration in amygdala-kindled rats (Wada et al., 1996; Kamei et al., 1998) and also decreased the duration of clonic convulsions, but not tonic ones, following electroconvulsions (Yokoyama et al., 1992). On the contrary, Borowicz et al. (2000) found no effect of L-histidine (histamine precursor; up to 2500 mg/kg) upon amygdala-kindled seizures in rats or pentylenetetrazole- and aminophylline-induced convulsions in mice. Moreover, α-fluoromethylhistidine, an inhibitor of histamine synthesis, showed a potent proconvulsant effect on clonic and convulsive coma phase in mice (Yokoyama et al., 1992). High doses of the centrally acting H_1 receptor antagonists, diphenhydramine and pyrilamine, were shown to potentiate chemically induced convulsions in mice (Fairbairn and Sturman, 1989).

At therapeutic dosages, many of the older classical H_1 receptor antagonists give rise to sedative side effects that have been attributed to occupancy of H_1 receptors in the CNS (Schwartz et al., 1981; Nicholson et al., 1991; Leurs et al., 1995). Most of the first generation H_1-antihistamines readily crosses the blood–brain barrier. Numerous clinical reports have shown that histamine H_1 receptor antagonists occasionally induced convulsions in children, especially in those of preschool ages (Wyngarden and Seevers, 1951; Schwartz and Patterson, 1978) and in adult epileptic patients (Churchull and Gammon, 1949). However, several compounds that penetrate poorly into the CNS and appear to be devoid of central depressant effects are now available. These second generation drugs include terfenadine, astemizole, loratadine, acrivastine and cetirizine.

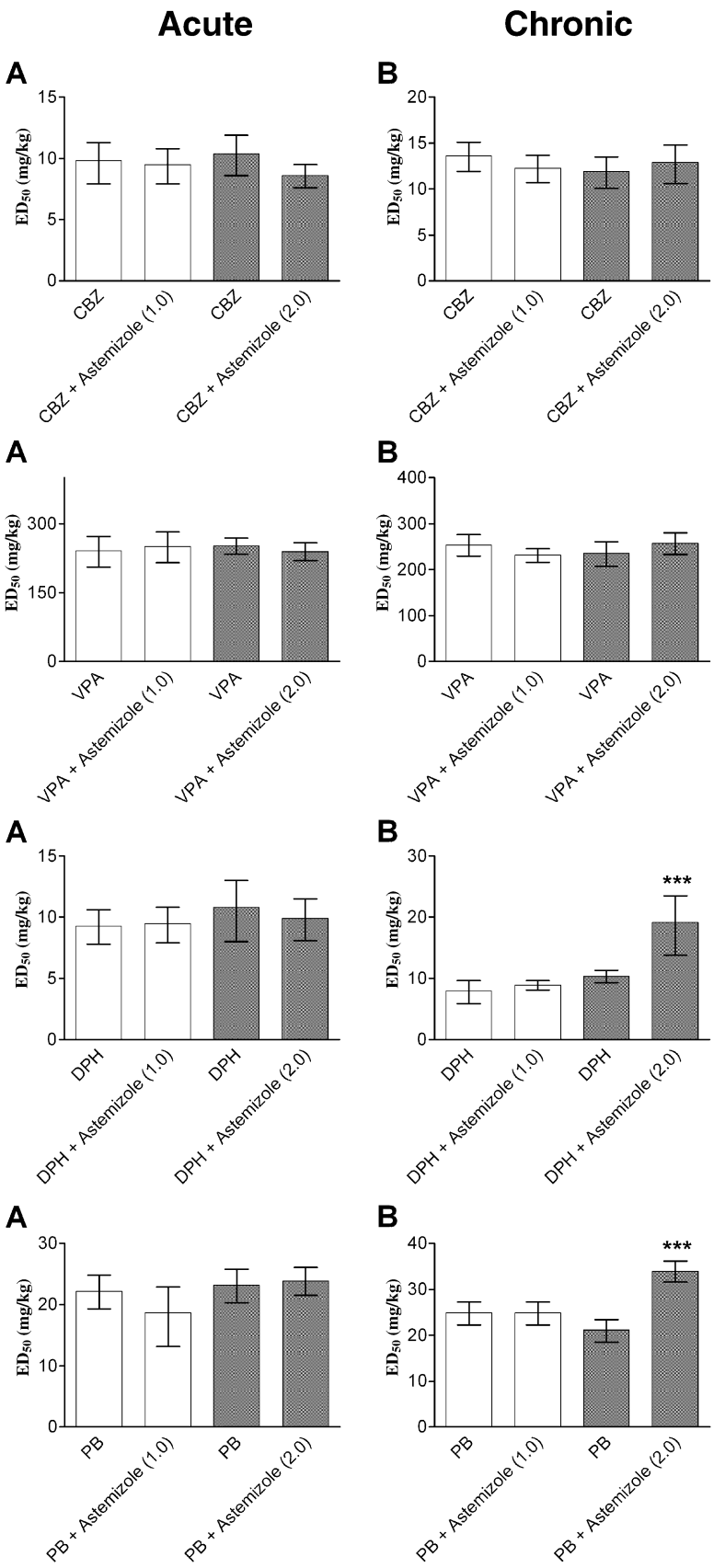
In the present study, we examined the influence of the second generation H_1 receptor antagonist, astemizole, administered acutely or once daily for 7 days, on the anticonvulsant activity of conventional antiepileptic drugs (AEDs) against MES-induced seizures in mice. Moreover, we evaluated effects of the tested drugs on long-term memory and motor coordination in mice. To define a possible involvement of pharmacokinetic mechanisms in the observed effects, the influence of astemizole on the free plasma levels and brain concentrations of the AEDs was also studied. Part of this study has already been published as a proceeding of 6th Conference on Progress in Ethiopathogenesis of Seizures (Świąder et al., 2001).

2. Materials and methods

2.1. Animals

The experiments were carried out on male Swiss mice weighing 20–25 g. The animals were housed in colony cages with free access to food (chow pellets) and tap water. The experimental temperature was 21±1 °C and mice were on a natural light–dark cycle. After 7 days of adaptation to laboratory conditions, the animals were randomly assigned to experimental groups (consisting of 8–12 animals). Each mouse was used only once. Local Bioethical Committee of Lublin approved all experimental procedures.

Fig. 2. Influence of astemizole (2 mg/kg ip in 1% Tween 80), given acutely or for 7 days, upon the protective activity of antiepileptic drugs (AEDs) against maximal electroshock-induced seizures in mice. Carbamazepine (CBZ) and valproate (VPA) were given intraperitoneally 30 min, phenobarbital (PB), 60 min and diphenylhydantoin (DPH), 120 min before testing. (A) Astemizole in a single dose was given intraperitoneally 30 min before testing. (B) Chronic treatment with astemizole (once daily intraperitoneally for 6 days), was also given on the seventh day, 30 min before testing. Astemizole 1 mg/kg+AEDs (white bars) or 2 mg/kg+AEDs (black bars) were statistically compared with the respective control groups (AEDs alone). At least four groups of mice, consisting of eight animals, were used to estimate each ED_{50} value (50% effective dose with 95% confidence limits). The data are ED_{50} values of AEDs, calculated and compared according to Litchfield and Wilcoxon (1949). ***P<.001.


Acute
Chronic
A
B
ED50 (mg/kg)
CBZ
CBZ + Astemizole (1.0)
CBZ + Astemizole (2.0)
VPA
VPA + Astemizole (1.0)
VPA + Astemizole (2.0)
DPH
DPH + Astemizole (1.0)
DPH + Astemizole (2.0)
PB
PB + Astemizole (1.0)
PB + Astemizole (2.0)
***

2.2. Drugs

The following drugs were used throughout the study: astemizole (Polfa, Warszawa, Poland) and four conventional AEDs: valproate magnesium (Dipromal, kindly donated by ICN Polfa Rzeszów, Poland), carbamazepine (Amizepin, Polfa, Starogard, Poland), diphenylhydantoin (Phenytoinum) and phenobarbital sodium (Luminalum Natrium, both drugs from Polfa). Phenobarbital and valproate magnesium were dissolved in distilled water, whilst astemizole, carbamazepine and diphenylhydantoin were suspended in a 1% solution of Tween 80 (Sigma, St. Louis, MO, USA). All drugs were administered intraperitoneally in a volume of 0.01 ml/g body weight. Pretreatment times for astemizole, valproate magnesium and carbamazepine were 30 min, phenobarbital, 60 min and diphenylhydantoin, 120 min prior to the tests. The doses of phenobarbital and valproate refer to their free forms.

2.3. Electroconvulsions

Electroconvulsions were produced using ear-clip electrodes and alternating current delivered by a Hugo Sachs (Type 221, Freiburg, Germany) generator, the stimulus duration being 0.2 s. Tonic hindlimb extension was taken as the endpoint. The convulsive threshold was evaluated as CS_{50}, which is the current strength (in mA) necessary to produce tonic hindlimb extension in 50% of the animals tested. To estimate the convulsive threshold, at least four groups of mice (eight animals per group) were challenged with electroshocks of various intensities. Subsequently, an intensity–response curve was calculated on the basis of the percentage of mice convulsing. The control group was injected with 1% Tween 80 whilst the remaining groups received different doses of astemizole. To evaluate the respective ED_{50} values (in mg/kg; effective doses of AEDs required to block the hindlimb tonic extension in 50% of the animals), mice pretreated with different doses of AEDs (with or without astemizole) were challenged with MES (25 mA). At least four groups of mice, consisting of eight animals, were used to estimate each ED_{50} value. A dose–effect curve was constructed, based on the percentage of mice protected, according to the method of Litchfield and Wilcoxon (1949). Control groups received AEDs alone +1% Tween 80 whilst the experimental groups were given AEDs+astemizole.

2.4. Chimney test

Motor impairment was evaluated with the chimney test of Boissier et al. (1960). In this test, animals had to climb backwards up a plastic tube (3-cm inner diameter, 25-cm length). Motor impairment was indicated by the inability of mice to climb backwards up the tube within 60 s. The animals were pretrained 24 h before treatment and those unable to perform the test were rejected from experimental groups. TD_{50} values (in mg/kg; toxic doses of AEDs required to produce motor impairment in 50% of the animals) for conventional AEDs alone or in combination with astemizole (administered at the maximal dose of 2 mg/kg) were evaluated, according to the method of Litchfield and Wilcoxon (1949).

2.5. Passive avoidance acquisition and retention testing

According to Venault et al. (1986), the step-through passive avoidance task may be applied as a measure of long-term memory acquisition. We used this test to compare the influence of astemizole and AEDs alone or in combinations on passive avoidance acquisition in mice. The animals were placed in an illuminated box (10×13×15 cm) connected to a larger (25×20×15 cm) dark compartment equipped with an electric grid floor. In this test, entry into the dark compartment was punished by an electric footshock (0.6 mA for 2 s; facilitation of acquisition). The AEDs or astemizole were given to animals on the first day, at times scheduled for the convulsive test, before the training session. The pretreated mice, that did not enter the dark compartment within 60 s, were excluded from the experiment. On the following day (24 h later), the same animals, without any treatment, were again placed in the illuminated box and the retention time was measured. The mice avoiding the dark compartment for longer than 180 s were regarded as remembering the task. Retention was expressed as the medians with 25 and 75 percentiles of at least 12 determinations.

2.6. Estimation of the free plasma levels and brain concentrations of AEDs

Plasma levels of AEDs were measured according to Czuczwar et al. (1989). The animals were given either one of the studied AEDs and 1% Tween 80 (control group) or combinations of the H_1 receptor antagonist with AEDs. Mice were decapitated at times scheduled for the convulsive test and blood samples of approximately 1 ml were collected into Eppendorf tubes. The whole brains were taken from the same animals at 4 °C, immediately, after decapitation. According to Borowicz et al. (1999) brains of mice were homogenized in TDx buffer (Abbott, Irving, TX, USA) in a proportion buffer/tissue 2:1 (v/w). Samples of blood and

Fig. 3. Influence of astemizole, injected singly or following a 7-day treatment, upon motor impairment produced by conventional antiepileptic drugs (AEDs). Carbamazepine (CBZ) and valproate (VPA) were given intraperitoneally 30 min, phenobarbital (PB), 60 min and diphenylhydantoin (DPH), 120 min before testing. (A) Astemizole in a single dose was given intraperitoneally 30 min before testing. (B) Astemizole following chronic intraperitoneally treatment for 6 days, was also injected on the seventh day, 30 min before testing. At least four groups of mice, consisting of eight animals, were used to estimate each TD_{50} value (50% toxic dose with 95% confidence limits). The data are TD_{50} values of AEDs, calculated and compared according to Litchfield and Wilcoxon (1949). ***P<.001.

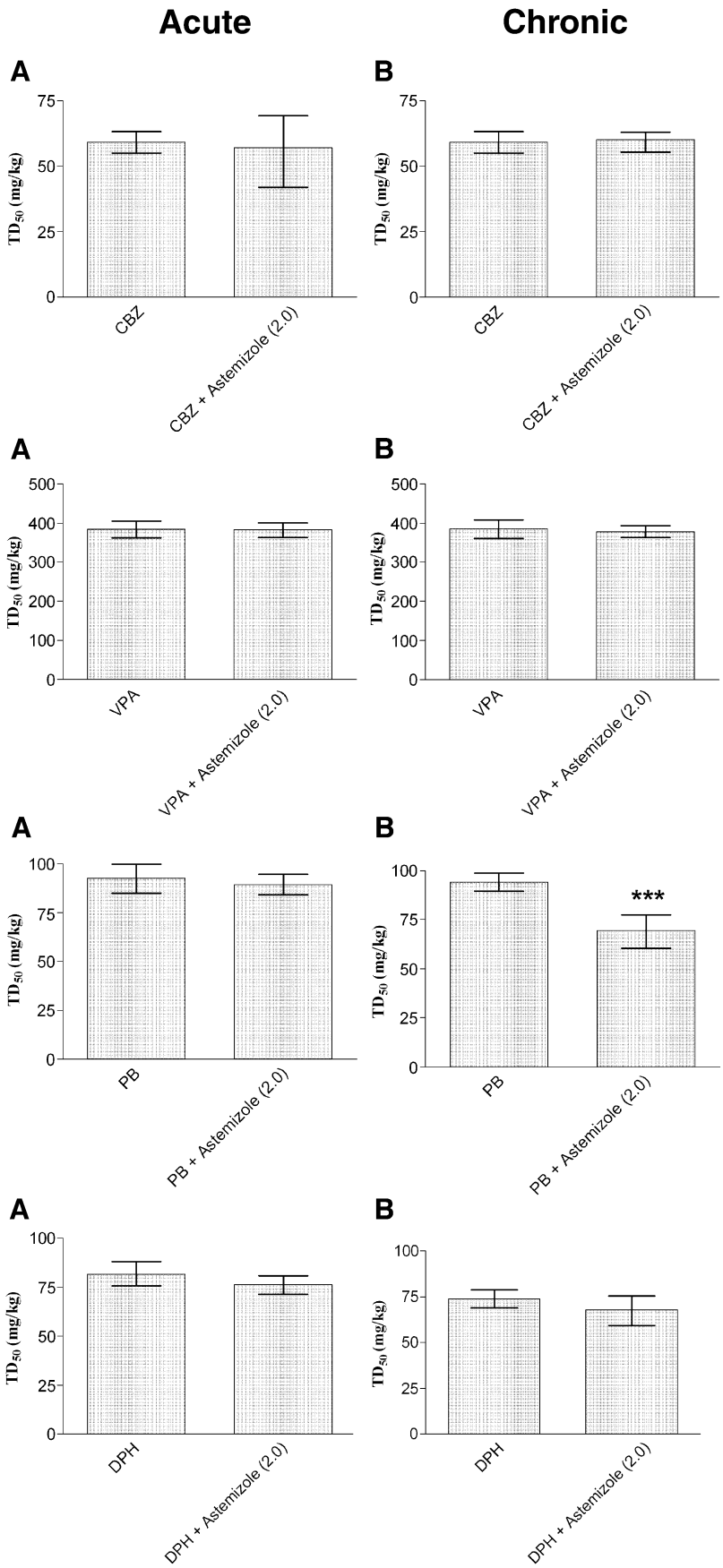

Acute
Chronic
A
B
TD50 (mg/kg)
CBZ
CBZ + Astemizole (2.0)
VPA
VPA + Astemizole (2.0)
PB
PB + Astemizole (2.0)
DPH
DPH + Astemizole (2.0)
***

Table 1
Effects of astemizole (after acute or chronic treatment) and antiepileptic drugs or their combinations on the retention time (s) in a passive avoidance task in mice

Drugs	Dosage (mg/kg)		Astemizole	Medians (25,75 percentiles)	
	Acute	Chronic		Acute	Chronic
Vehicle	–	–	–	180 (180,180)	180 (180,180)
Astemizole	–	–	2	180 (148,180)	180 (47.5,180)
Carbamazepine	10.4	11.9	–	180 (124.5,180)	180 (174,180)
Carbamazepine	8.6	12.9	–	180 (144,180)	180 (180,180)
Carbamazepine	8.6	12.9	2	180 (146,180)	180 (119,180)
Valproate	252	235	–	133 (90.5,180)	141 (89,180)
Valproate	240	257	–	156 (97,180)	139 (41,180)
Valproate	240	257	2	131 (89,180)	99 (37,180)
Phenobarbital	23.2	21.1	–	149 (103,180)	180 (161,180)
Phenobarbital	23.9	34.0	–	152 (104,180)	124 (76,150)*
Phenobarbital	23.9	34.0	2	134 (97.5,180)	120 (48,159)**
Diphenylhydantoin	10.8	10.4	–	180 (136,180)	180 (127,180)
Diphenylhydantoin	9.9	19.2	–	180 (157,180)	103 (59,176)*,#
Diphenylhydantoin	9.9	19.2	2	180 (155,180)	109 (50,171)*,#

Presented values are medians with 25 and 75 percentiles of 12 determinations. The retention was expressed as a period time (s) in which the animals avoided the dark compartment. Carbamazepine and valproate were given intraperitoneally 30 min, phenobarbital, 60 min and diphenylhydantoin, 120 min before testing. Astemizole in a single dose was given intraperitoneally 30 min before testing. Chronic treatment with astemizole ended on the seventh day, 30 min before testing. The results obtained from the passive avoidance task were statistically verified by the Kruskal–Wallis nonparametric ANOVA test followed by the Dunn's post hoc test.

* $P<.05$.

** $P<.01$ vs. vehicle group.

\# $P<.05$ vs. diphenylhydantoin at the dose of 10.4 mg/kg.

brain were centrifuged and only plasma samples were pipetted into a micropartition system, MPS-1 (Amicon, Danvers, MA, USA). Again, the samples were centrifuged and the free plasma and brain levels of investigated groups were determined by immunofluorescence, using an Abbott TDx analyzer (Abbott, Irving, TX, USA). The plasma or brain levels of AEDs were expressed in μg/ml of plasma or μg/g of wet brain tissue as means±S.D. of at least eight determinations.

2.7. Treatment protocol

Experiments were carried out after acute or chronic treatments with astemizole.

1. Acute study: animals were injected with a single dose of the H_1 receptor antagonist and one of the AEDs at the time prior to the tests mentioned above. Conventional AEDs were tested at the time of their peak anticonvulsant activity, according to our previously published studies (Czuczwar et al., 1990; Gasior et al., 1996), whilst the maximal time of activity of astemizole was determined experimentally.
2. Chronic study: once a day (between 8:00 and 10:00 a.m.) mice were injected as follows: Group 1, 1% Tween 80 for 6 days (control group); Group 2, astemizole for 6 days. On the 7th day, mice from both groups received one of the conventional AEDs and vehicle or astemizole, respectively, just like in the acute study.

The CS_{50}s of astemizole, ED_{50}s and TD_{50}s of AEDs alone or in combination with astemizole were calculated and statistically analyzed. The effects of the acute or chronic H_1 receptor antagonist on the long-term memory and brain or free plasma levels of AEDs were also evaluated in both experimental protocols.

2.8. Statistics

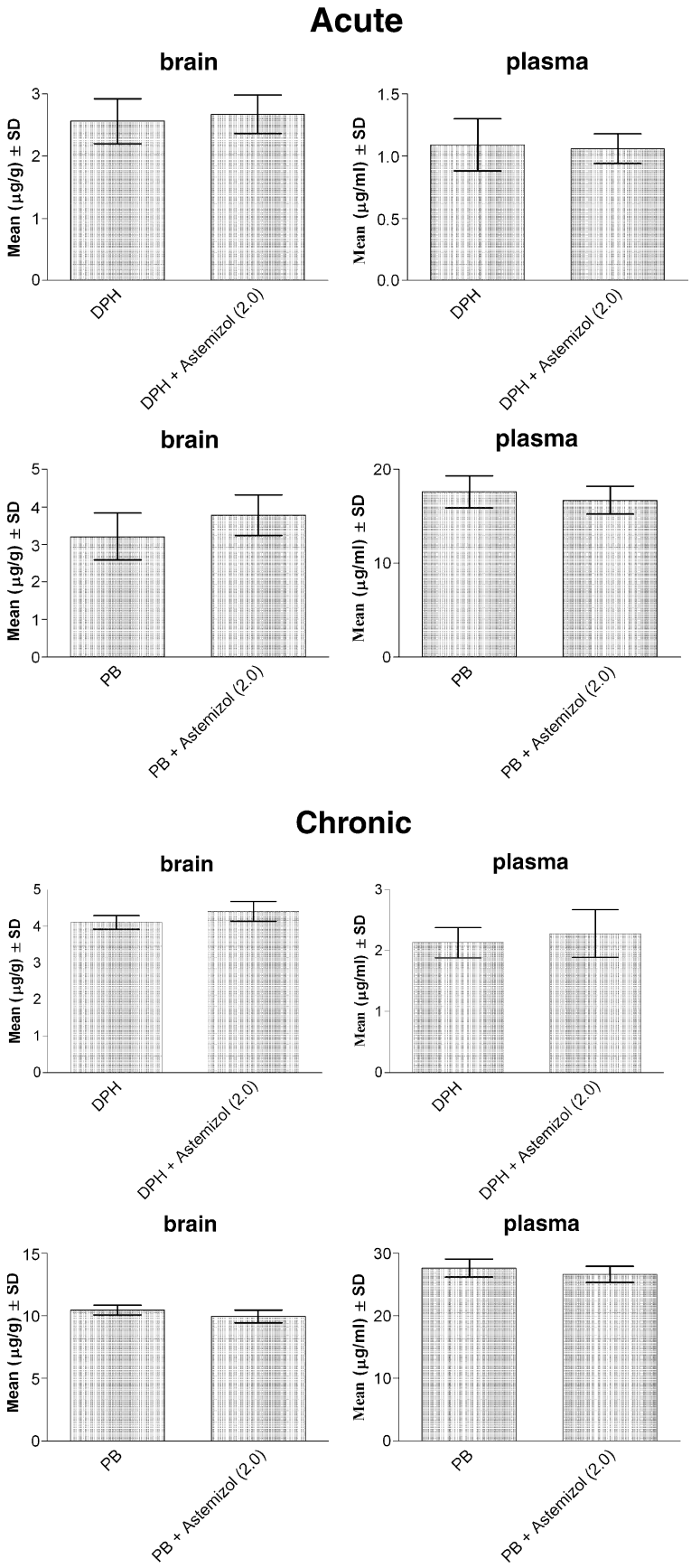
CS_{50}, ED_{50} and TD_{50} values and their statistical comparisons were calculated by computer probit analysis according to Litchfield and Wilcoxon (1949). The results from the passive avoidance task were statistically verified with the Kruskal–Wallis nonparametric ANOVA test followed by the Dunn's post hoc test. Unpaired Student's *t* test was used for the statistical evaluation of the free plasma and brain levels of AEDs.

3. Results

3.1. Effects of astemizole given acutely or after 7-day treatment on the electroconvulsive threshold

Astemizole (2.0, 3.0, 4.0, 6.0 mg/kg ip) administered acutely 30 min before the test, significantly reduced the electroconvulsive threshold from 5.8 to 4.6, 4.6, 4.3 and 4.6

Fig. 4. Influence of acute or chronic astemizole, on the free plasma levels or brain concentrations of AEDs. Phenobarbital (PB) was given intraperitoneally 60 min and diphenylhydantoin (DPH), 120 min before testing. Astemizole was given in a single dose intraperitoneally, 30 min before testing or after chronic treatment (once daily), on the seventh day, 30 min before testing. Presented values are the means in μg/ml of plasma±S.D. or μg/g wet brain tissue of eight mice. Blood and brain samples were taken at times scheduled for the convulsive test. Unpaired Student's *t* test was used for statistical evaluation of the data.


Acute
brain
plasma
Mean (µg/g) ± SD
Mean (µg/ml) ± SD
DPH
DPH + Astemizol (2.0)
PB
PB + Astemizol (2.0)
Chronic

mA, respectively. No effect was observed when astemizole was used at a lower dose of 1 mg/kg (Fig. 1).

Astemizole (2 mg/kg) given for 7 days, markedly decreased the electroconvulsive threshold, being without effect on this parameter at 1 mg/kg (Fig. 1).

3.2. Influence of acute or chronic astemizole on the protective activity of AEDs against MES-induced seizures in mice

Astemizole (up to 2 mg/kg), administered acutely, did not affect the antielectroshock activity of conventional AEDs. The ED50 values of carbamazepine (10.4 mg/kg), valproate (252 mg/kg), phenobarbital (23.2 mg/kg) or diphenylhydantoin (10.8 mg/kg) were not significantly modified during astemizole (2 mg/kg) coadministration. Also, astemizole, following the 7-day treatment, administered at the subthreshold dose of 1 mg/kg, did not affect the anticonvulsant activity of conventional AEDs either, whilst at the lowest effective dose of 2 mg/kg, it significantly diminished the antiseizure efficacy of phenobarbital and diphenylhydantoin. The ED_{50} value of phenobarbital was raised from 21.1 to 34.0 mg/kg. Similarly, the ED_{50} of diphenylhydantoin was elevated from 10.4 to 19.2 mg/kg. On the other hand, astemizole (2 mg/kg), given for 7 days, did not influence the protective action of valproate (235 mg/kg) or carbamazepine (11.9 mg/kg) against MES (Fig. 2).

3.3. Chimney test

Astemizole, in a single dose of 2 mg/kg, did not affect TD_{50} values of AEDs. Also, the repeated treatment with this drug at 2 mg/kg, did not influence the performance of mice in the chimney test when combined with valproate, carbamazepine or diphenylhydantoin. In contrast, astemizole (2 mg/kg) lowered the TD_{50} of phenobarbital from 94.3 to 69.5 mg/kg (Fig. 3).

3.4. Dark avoidance task

Neither astemizole (2 mg/kg), administered acutely alone nor combined with carbamazepine (8.6 mg/kg), valproate (240 mg/kg), diphenylhydantoin (9.9 mg/kg) or phenobarbital (23.9 mg/kg), which were given in their ED_{50} values against MES, impaired long-term memory in mice. Also these AEDs injected alone did not affect the memorizing processes in mice. Similarly, astemizole (2 mg/kg) administered for 7 days, alone or combined with carbamazepine and valproate, providing a 50% protection against electroconvulsions, did not affect the performance of mice in the passive avoidance task (Table 1). On the contrary, astemizole impaired memory performance when coadministered with phenobarbital or diphenylhydantoin when compared to the vehicle-pretreated control group. However, these AEDs (phenobarbital at 34 and diphenylhydantoin at 19.2 mg/kg) also exerted similar effects when given alone (Table 1). In addition, a significant disturbance was noted for diphenylhydantoin alone (19.2 mg/kg) or this drug coadministered with astemizole (2 mg/kg) in comparison with diphenylhydantoin alone at the dose of 10.4 mg/kg (Table 1).

3.5. Influence of astemizole on the free plasma and brain levels of AEDs

Astemizole (2 mg/kg), given acutely or chronically, did not affect the free plasma levels or brain concentrations of phenobarbital or diphenylhydantoin (Fig. 4).

4. Discussion

Around 30% of epileptic patients are not adequately controlled with AEDs in the form of either mono- or polytherapy (Deckers et al., 2000). At least in some of them, the therapeutic failure may result from the use of medications for other than epilepsy reasons. So far for example, methylxanthine derivatives (theophylline, pentoxifylline or caffeine) have been documented to sharply decrease the protective potential of conventional AEDs against MES-induced convulsions in mice (Czuczwar et al., 1990; Gasior et al., 1996). Interestingly, chronic caffeine was much more potent in this respect than the acute administration of the methylxanthine (Gasior et al., 1996). Consequently, other groups of drugs, especially those decreasing the convulsive threshold, need to be evaluated for their possible negative interactions with AEDs.

The results from these experiments indicate that astemizole reduced the threshold for electroconvulsions and at the lowest effective dose of 2 mg/kg, decreased the anticonvulsant activity of phenobarbital and diphenylhydantoin, but not that of valproate and carbamazepine. It is noteworthy that astemizole diminished the anticonvulsant properties of conventional AEDs only following 7-day treatment, being without effect upon the ED_{50} values of the AEDs when given acutely. Furthermore, astemizole did not affect the free plasma and brain concentrations of conventional AEDs, so the possibility of pharmacokinetic events may be excluded.

It was previously documented that classical H_1 receptor antagonists had proconvulsant effect in various models of epilepsy (Scherkl et al., 1991a; Yokoyama et al., 1992, 1993; Świąder et al., 1999). Moreover, L-histidine or metoprine, which elevate brain histamine levels, inhibited MES in rats (Duch et al., 1978, 1980) and increased the threshold for pentylenetetrazole-induced seizures in mice (Scherkl et al., 1991b). Furthermore, the second generation of H_1 receptor antagonists (e.g., astemizole, terfenadine) was completely ineffective in any convulsive tests (Scherkl et al., 1991a; Yokoyama et al., 1993). Kamei et al. (2000) have shown that the epileptogenic property of pyrilamine was more potent than that of chlorpheniramine or diphenhydramine, in contrast to the second generation of H_1 receptor antagonists,

loratadine and ebastine, which did not induce detectable epileptogenic activity.

Dux et al. (1987) reported that H_1 and H_2 receptor antagonists changed the permeability of blood–brain barrier and thereby their own distribution in the histaminergic neurons. Similarly, 7-day administration of astemizole could improve its penetration via the blood–brain barrier, leading to the reduced activity of some AEDs. It is noteworthy that in our studies astemizole was given at the maximal dose of 2 mg/kg, which was to 10-fold higher than that in the experiments of Scherkl et al. (1991b) but also in the same range of dosage as in studies of Yokoyama et al. (1993). Fairly high doses of histamine or histamine receptor antagonists may also affect the neuronal uptake or turnover rates of other monoamines (Tuomisto and Tuomisto, 1980; Shishido et al., 1991). It is necessary to consider effects of astemizole on other neurotransmitter system (noradrenergic, serotoninergic or cholinergic) since such effects were evident for a number of histamine H_1 receptor antagonists (Nowak, 1980; Philippu et al., 1984). However, Awouters et al. (1983) have found that astemizole, as a very specific H_1 receptor antagonist, is devoid of antagonistic properties against serotonin, muscarine, dopamine or catecholamines, and of a series of nonspecific activities. This may speak for somewhat specific effects of astemizole upon diphenylhydantoin and phenobarbital. However, some peripheral actions of astemizole with possible consequences on central regulatory mechanisms cannot be entirely excluded.

Moreover, the chronic and acute administration of astemizole, at the dose of 2 mg/kg, alone or in combinations with conventional AEDs was devoid of any significant adverse effects in the passive avoidance task or chimney test, except of the 7-day treatment with the drug. In this case, astemizole impaired motor coordination when coadministered with phenobarbital or long-term memory when combined with phenobarbital or diphenylhydantoin. However, these AEDs alone also caused memory impairment in mice. This indicates that the final memory impairment cannot be actually ascribed to astemizole since the effects of these AEDs alone or combined with the H_1 receptor antagonist were not statistically different. It is well known that compounds possessing atropinic properties considerably impair memory tasks in mice (Malmberg-Aiello et al., 2000). Considering that astemizole is devoid of the atropinic effect, it is not surprising that the drug did not influence memory task in the present study.

Our findings might be important from a clinical point of view because the second generation antihistamines are the drugs widely used in allergic diseases. It is important to underline that especially in pediatric patients (e.g., skin diseases like urticaria) these drugs are commonly prescribed. Finally, the present data indicate that astemizole needs to be used with caution in patients of risk, especially children in preschool age and epileptic patients, especially treated with diphenylhydantoin or phenobarbital.

Acknowledgements

This study was supported by the Grant No. P05A 044 18 from the State Committee for Scientific Research, Warszawa, Poland.

References

Awouters FHL, Niemegeers CJE, Janssen PAJ. Pharmacology of the specific histamine H1-antagonist astemizole. Arzneim-Forsch 1983;33:3381–8.

Boissier J-R, Tardy J, Diverres J-C. Une nouvelle methode simple pour explorer l'action 'tranquilisante': Le test de la cheminee. Med Exp 1960;3:81–4.

Borowicz KK, Kamiński R, Gąsior M, Kleinrok Z, Czuczwar SJ. Influence of melatonin upon the protective action of conventional anti-epileptic drugs against maximal electroshock in mice. Eur Neuropsychopharmacol 1999;9:185–90.

Borowicz KK, Świąder M, Kamiński R, Kuźniar H, Kleinrok Z, Czuczwar SJ. Two essential amino acids, L-lysine and L-histidine, in five types of experimental seizures. Pol J Pharmacol 2000;52:345–52.

Churchull JA, Gammon GD. The effect of antihistaminic drugs on convulsive seizures. J Am Med Assoc 1949;141:18–21.

Czuczwar SJ, Janusz W, Szczepanik B, Wamil A, Kleinrok Z. Effect of aminophylline upon the protective activity of common antiepileptic drugs and their plasma levels in mice. Neurosci Res 1989;6:470–4.

Czuczwar SJ, Gasior M, Janusz W, Szczepanik B, Wlodarczyk D, Kleinrok Z. Influence of different methylxanthines on the anticonvulsant action of common antiepileptic drugs in mice. Epilepsia 1990;31(3):318–3.

Deckers CLP, Czuczwar SJ, Hekster YA, Keyser A, Kubova H, Meinardi H, et al. Selection of antiepileptic drug polytherapy based on mechanism of action: the evidence reviewed. Epilepsia 2000;41:1364–74.

Duch DS, Bowers SW, Nichol CA. Elevation of brain histamine levels by diaminopyrimidine inhibitors of histamine *N*-methyltransferase. Biochem Pharmacol 1978;27:1507–9.

Duch DS, Edelstein MP, Nichol CA. Inhibition of histamine-metabolizing enzymes and elevation of histamine levels in tissues by lipid-soluble anticancer folate antagonists. Mol Pharmacol 1980;18:100–4.

Dux E, Temesvári P, Szedahelyi P, Nagy A, Kovacs J, Joó F. Protective effect of antihistamines on cerebral oedema induced by experimental pneumothorax in newborn piglets. Neuroscience 1987;22:317–21.

Fairbairn S, Sturman G. The influence of central histaminergic modification on the susceptibility of mice to chemically-induced seizures [abstract]. Br J Pharmacol 1989;97:317 [suppl.].

Gasior M, Borowicz K, Buszewicz G, Kleinrok Z, Czuczwar SJ. Anticonvulsant activity of phenobarbital and valproate against maximal electroshock in mice during chronic treatment with caffeine and caffeine discontinuation. Epilepsia 1996;37(3):262–8.

Gerald MC, Richter NA. Studies on the effects of histaminergic agents on seizure susceptibility in mice. Psychopharmacologia 1976;46:277–82.

Kamei C, Ishizawa K, Kakinoki H, Fukunaga M. Histaminergic mechanisms in amygdaloid-kindled seizures in rats. Epilepsy Res 1998;30:187–94.

Kamei C, Ohuchi M, Sugimoto Y, Okuma C. Mechanism responsible for epileptogenic activity by first-generation H1-antagonists in rats. Brain Res 2000;887:183–6.

Leurs R, Smit MJ, Timmerman H. Molecular pharmacological aspects of histamine receptors. Pharmacol Ther 1995;66:413–63.

Litchfield JT, Wilcoxon F. A simplified method of evaluating dose–effect experiments. J Pharmacol Exp Ther 1949;96:99–113.

Malmberg-Aiello P, Ipponi A, Bartolini A, Schunack W. Antiamnesic effect of metoprine and of selective histamine H(1) receptor agonists in a modified mouse passive avoidance test. Neurosci Lett 2000;288:1–4.

Nicholson AN, Pascoe PA, Turner C, Ganellin CR, Casay AF, Mercer AD. Sedation and histamine H1-receptor antagonism: studies in man with

the enantiomers of chlorpheniramine and dimethindene. Br J Pharmacol 1991;104:270–6.

Nowak JZ. Effects of histamine H1- and H2-receptor antagonists on dopamine, noradrenaline and serotonin systems in rat brain. Pol J Pharmacol Pharm 1980;32:451–61.

Onodera K, Tuomisto L, Tacke U, Airaksinen M. Strain differences in regional brain histamine levels between genetically epilepsy-prone and resistant rats. Methods Find Exp Clin Pharmacol 1992;14:13–6.

Philippu A, Bald M, Kraus A, Dietl H. In vivo release by histamine agonists and antagonists of endogenous catecholamines in the cat hypothalamus. Naunyn-Schmiedeberg's Arch Pharmacol 1984;326:116–23.

Scherkl R, Hashem A, Frey HH. Importance of histamine seizure susceptibility. In: Timmerman H, van der Groot H, editors. New perspectives in histamine research. Basel: Birkhäuser; 1991a. p. 85–9.

Scherkl R, Hashem A, Frey H. Histamine in brain—its role in regulation of seizure susceptibility. Epilepsy Res 1991b;10:111–8.

Schwartz JF, Patterson JH. Toxic encephalopathy related to antihistamine-barbiturate antiemetic medication. Am J Dis Child 1978;32:37–9.

Schwartz JC, Garbarg M, Quach TT. Histamine receptors in brain, a target for tricyclic antidepressants. Trends Pharmacol Sci 1981;2:122–5.

Schwartz JC, Arrang JM, Garbarg M, Pollard H, Ruat M. Histaminergic transmission in the mammalian brain. Physiol Rev 1991;71:1–51.

Shishido S, Oishi R, Saeki K. In vivo effect of some histamine H1-receptor antagonists on monoamine metabolism in mouse brain. Naunyn-Schmiedeberg's Arch Pharmacol 1991;343:185–9.

Świąder M, Kalisz A, Porąbiak J, Kleinrok Z, Czuczwar SJ. Influence of antazoline on the anticonvulsant activity of conventional antiepileptic drugs against maximal electroshock-induced seizures in mice [abstract]. Pol J Pharmacol 1999;51:104.

Świąder M, Chwalczuk K, Wielosz M, Czuczwar SJ. Influence of chronic treatment with H1 receptor antagonists on the anticonvulsant activity of antiepileptic drugs. Pol J Pharmacol 2001;53:93–6.

Tuomisto L, Tacke U. Is histamine an anticonvulsive inhibitory transmitter? Neuropharmacology 1986;25:955–8.

Tuomisto J, Tuomisto L. Effects of histamine and histamine antagonists on the uptake and release of catecholamines and 5-HT in brain synaptosomes. Med Biol 1980;58:33–7.

Venault P, Chapouthier P, de Carvallo LP, Simiand J, Morre M, Dodd RH, et al. Benzodiazepines impair and beta-carbolines enhance performance in learning and memory tasks. Nature 1986;321:864–866.

Wada H, Inagaki N, Yamatodani A, Watanabe T. Is the histaminergic neuron system a regulatory center for whole-brain activity? Trends Neurosci 1991;14:415–9.

Wada Y, Shiraishi J, Nakamura M, Koshino Y. Biphasic action of the histamine precursor L-histidine in the rat kindling model of epilepsy. Neurosci Lett 1996;204:205–8.

Wyngarden JB, Seevers MH. The toxic effects of antihistaminic drugs. J Am Med Assoc 1951;145:277–82.

Yokoyama H, Onodera K, Maeyama K, Yanai K, Iinuma K, Tuomisto L, et al. Histamine levels and clonic convulsions of electrically-induced seizure in mice: the effects of α-fluoromethylhistidine and metoprine. Naunyn-Schmiedeberg's Arch Pharmacol 1992;346:40–5.

Yokoyama H, Onodera K, Iinuma K, Watanabe T. Proconvulsive effects of histamine H1-antagonists on electrically-induced seizure in developing mice. Psychopharmacology 1993;112:199–203.