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# *Felbamate reduces exploratory and spontaneous locomotor activities in mice*

Polytherapy in epilepsy is advocated as a preferred treatment in patients with intractable seizures (4). In such cases, the rationale for combining some antiepileptic drugs (AEDs) is usually based upon the presumptions concerning two aspects of the efficacious treatment in epilepsy. The first aspect is directly related to the anticonvulsant activity of combined drugs, whilst the second one takes into consideration the side-effect profile of co-administered AEDs (2, 5).

Relatively recently, the electronically-monitored locomotor activity test has been accepted as an eligible and sophisticated model for evaluating a potential risk of neurotoxic adverse-effects produced by drugs with respect to the impairment of locomotor activity in animals (3, 9). Therefore, it can be considered as an experimental model useful for assessing neurotoxic effects produced by AEDs either alone or in combinations in terms of spontaneous and exploratory activities in animals.

The aim of this study was to evaluate the effect of felbamate (FBM) and lamotrigine (LTG) – two newer AEDs, clinically used in patients with generalized tonic-clonic convulsions, and partial seizures with or without secondary generalization (1), on the exploratory and spontaneous locomotor activities in mice. The alterations in locomotor functioning of animals following a single exposure to FBM alone or combined with LTG would allow us to determine the adverse-effect profile of these AEDs.

## MATERIAL AND METHODS

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. All procedures listed were approved by Local Ethics Committee at the Medical University of Lublin.

Drugs. The following drugs were used in this study: FBM (Schering Plough, Levallois Perret, France) and LTG (Glaxo Wellcome, Kent, UK). Both AEDs were suspended in a 1% solution of Tween 80 (Sigma, St. Louis, MO, USA) in distilled water and administered intraperitoneally (i.p.) in a volume of 0.1 ml/g body weigh, 60 min prior to the test.

Locomotor activity monitoring. Locomotor activity of animals was assessed with a Digiscan Animal Activity Monitor System (Omnitech Electronics, Columbus, OH, USA). Each monitor consisted of a 41 x 41 x 32 cm Plexiglas open field box with a grid of infrared beams mounted horizontally every 2.5 cm and vertically every 4.5 cm. Photocells located on the wall directly

opposite each photo-beam were activated when the animal interrupted the beam. Each box was partitioned with acrylic cross into four (20 x 20 x 32 cm) quadrants. Mice were tested in the opposite quadrants of each unit (i.e. two mice per box). The photocells of each activity box were connected to the Digiscan analyzer that transmitted the number of beam breaks (activity data) to a computer. During operation, the pattern of beam interruptions was recorded and analyzed by IBM-PC compatible computer. Interruption of any beam was recorded as an activity score. All activity data were collected during two consecutive 15-min periods. Cumulative counts were compiled and downloaded every 15 min into the data collection software, which organized these counts into different motor indices. Two representative motor indices were analyzed as follows: 1) total distance traveled in cm – measuring the amount of forward activity of animals, and 2) rearing activity – measuring the total number of vertical photo-beam interruptions within the sample period.

Experimental design. In this experiment, the mice were not habituated to the test apparatus; therefore, the test procedure consisted of two independent, consecutive measures. After injection of vehicle or the respective AED doses, the mice were placed in the centre of individual cages and recording of their locomotor activities started throughout two consecutive 15 min-intervals. The first measure of animals' activity is the rate of habituation to a novel environment. Thus, during prolonged exposure to a new environment, animals typically spend progressively less time in movements and exploration. So, the second measure is considered as the rate of spontaneous activity of mice.

Statistics. Data from the locomotor activity test were statistically evaluated with the analysis of variance (ANOVA) followed by Bonferroni's post-hoc test for multiple comparisons.

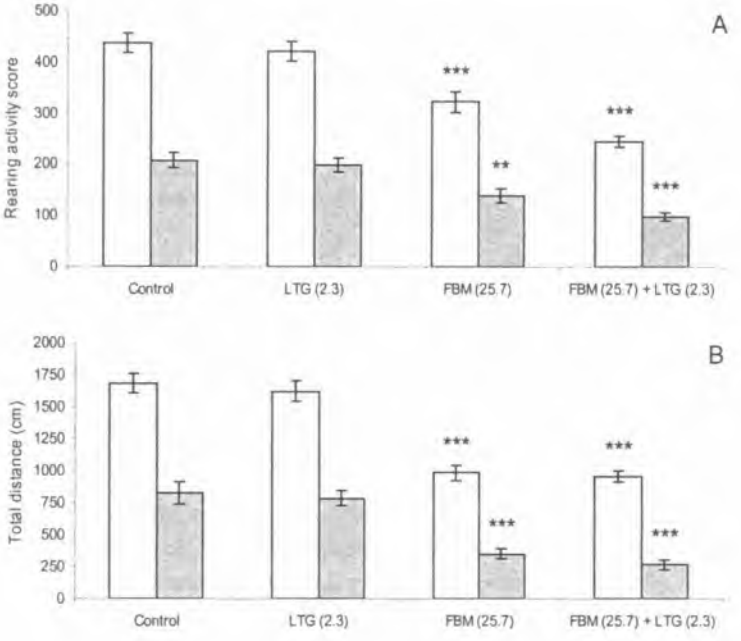
## RESULTS

### EXPLORATORY LOCOMOTOR ACTIVITY TESTING

FBM (at 25.7 mg/kg) combined with LTG (2.3 mg/kg) markedly reduced rearing activity scores in mice from 437 to 245 ($p<0.001$) (Fig. 1A). Likewise, FBM (25.7 mg/kg) injected separately evoked a significant reduction in rearing activity scores from 437 to 323 ($p<0.01$) (Fig. 1A). Again, LTG (2.3 mg/kg) administered alone did not affect rearing activity scores in animals tested (Fig. 1A). Moreover, statistical analysis of data revealed that FBM (25.7 mg/kg) injected singly or in combination with LTG (2.3 mg/kg) significantly shortened the total distance traveled by animals from 1684 to 984 and 957 cm, respectively ($p<0.001$) (Fig. 1B). In contrast, LTG (2.3 mg/kg) administered alone had no impact on total distance traveled by the animals (Fig. 1B).

### SPONTANEOUS LOCOMOTOR ACTIVITY TESTING

FBM combined with LTG considerably reduced rearing activity scores in mice from 208 to 98 ($p<0.001$) (Fig. 1A). Similarly, FBM injected singly evoked a significant reduction in rearing activity scores from 208 to 139 ($p<0.01$) (Fig. 1A). Only, LTG (2.3 mg/kg) administered separately did not alter this parameter in animals tested (Fig. 1A). Additionally, it was found that FBM (25.7 mg/kg) in combination with LTG (2.3 mg/kg) significantly reduced the total distance traveled by animals from 827 to 268 cm ($p<0.001$) (Fig. 1B). The same effect was observed for FBM (25.7 mg/kg) administered alone that markedly shortened total distance traveled by animals from 827 to 352 cm ($p<0.001$) (Fig. 1B). In contrast, LTG (2.3 mg/kg) administered separately had no impact on total distance traveled by the mice (Fig. 1B).




Figures 1A–1B. Influence of felbamate alone or combined with lamotrigine on the exploratory and spontaneous locomotor activities in mice

Figure A represents the effect of felbamate (FBM) and lamotrigine (LTG) administered alone or in combination on rearing activity in mice, whereas Figure B illustrates the effect of FBM and LTG on total distance traveled by animals in the exploratory and spontaneous locomotor activity test. White columns represent on graph the exploratory locomotor activity of the animals tested (as means ± SEM, as the error bars) evaluated during the first period of time (0-15 min) – habituation to a novel environment. The grey columns represent on graph the means ± SEM (as the error bars) of spontaneous locomotor activity in animals, evaluated during the second period of time (15-30 min). Statistical analysis of data with one-way ANOVA followed by post-hoc Bonferroni's test revealed a significant reduction in both exploratory and spontaneous locomotor activities as compared to the control (vehicle-treated animals) values at $^{**}p<0.01$, and $^{***}p<0.001$, respectively

## DISCUSSION

Our findings indicate clearly that FBM administered alone or combined with LTG markedly reduced exploratory and spontaneous locomotor activities in mice with relation to all the examined parameters. In fact, FBM at a constant dose of 25.7 mg/kg decreased the ambulatory (7) and rearing activities as well as shortening the total distance traveled by animals in both, the exploratory and spontaneous locomotor activities in mice. In contrast, neither the exploratory nor spontaneous locomotor activities of animals were altered after the injection of LTG alone. It is noteworthy that the doses of both AEDs used in this study were based on our previous experiment, evaluating the effects of FBM and LTG against maximal electroshock-induced seizures in mice (6).

The observed reduction in exploratory and spontaneous locomotor activities following the i.p. administration of FBM, when compared to control (vehicle-treated animals), provides evidence that the drug may not be useful in the clinical setting. It is highly likely that FBM may reduce activity in patients receiving FBM, since a high positive correlation exists between the adverse-effect profiles of AEDs evaluated in rodents and humans (8).

It should be highlighted that the electronically-monitored locomotor activity test in rodents seems to be a good screening allowing preselection of drugs affecting CNS and changing locomotor functioning in experimental animals.

## CONCLUSIONS

1. The utmost caution is advised when administering FBM to epileptic patients since the drug remarkably reduced the exploratory and spontaneous locomotor activities in animals.
2. Lack of any significant changes in locomotor activity of animals injected with LTG alone, during the exploratory and spontaneous activities testing, is of some clinical importance, worthy of further consideration.

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## SUMMARY

This study examines the effects of felbamate administered either alone or in combination with lamotrigine on the exploratory and spontaneous locomotion in mice. Locomotor activities were monitored electronically, evaluating the rearing (vertical) activity and total distance traveled by animals

within two 15-min periods. Results indicate that felbamate (25.7 mg/kg) administered separately or combined with lamotrigine (2.3 mg/kg) considerably reduced all the investigated parameters of the exploratory and spontaneous locomotor activities in mice. In contrast, lamotrigine (2.3 mg/kg) administered alone had no impact on the rearing activity and total distance traveled by animals with respect to the exploratory and spontaneous locomotor activities. In conclusion, since felbamate reduced locomotor activities in experimental animals, utmost caution is advised when combining the drug with other antiepileptic drugs in clinical practice.

Felbamat zmniejsza poznawczą oraz spontaniczną aktywność ruchową u myszy

Praca ocenia wpływ felbamatu podawanego osobno jak i w kombinacji z lamotryginą, na poznawczą i spontaniczną aktywność ruchową u myszy. Aktywność ruchową monitorowano elektronicznie, oceniając ruchliwość pionową oraz całkowity dystans pokonany przez zwierzęta w ciągu 2 okresów po 15 min. Wyniki wykazują, że felbamat (25,7 mg/kg) podawany osobno lub w kombinacji z lamotryginą (2,3 mg/kg) znacząco zmniejszał wszystkie oceniane parametry aktywności poznawczej i spontanicznej u myszy. Przeciwnie, lamotrygina (2,3 mg/kg) podawana osobno nie miała żadnego wpływu na ruchliwość pionową oraz całkowity dystans pokonany przez zwierzęta w odniesieniu do aktywności poznawczej i spontanicznej. Wnioskując, skoro felbamat zmniejszał aktywność motoryczną u zwierząt doświadczalnych, zaleca się znaczną ostrożność podczas łącznego podawania leku z innymi lekami przeciwpadaczkowymi w warunkach klinicznych.