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European Journal of Pharmacology 481 (2003) 67–74
www.elsevier.com/locate/ejphar

Anxiolytic effects of the novel anti-epileptic drug levetiracetam in the elevated plus-maze test in the rat

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Received 12 May 2003; received in revised form 28 August 2003; accepted 3 September 2003

Abstract

There is clinical evidence of anxiolytic action of several anti-epileptic drugs. We evaluated the effects of levetiracetam (Keppra™), a new generation anti-epileptic drug, in the plus-maze animal test for anxiolytic activity. Levetiracetam at 17 and 54 mg/kg intraperitoneally (i.p.) was without effect when tested in naive rats. A modified version of the test was subsequently used in which open-arm exploration was decreased by exposure of the rats to a four-open-arm maze 24 h prior to drug treatment and testing. Under these conditions of enhanced anxiety, levetiracetam, 5.4 to 54 mg/kg, dose-dependently increased open-arm exploration. Chlordiazepoxide 5 mg/kg had similar effects although buspirone 0.1 to 1.0 mg/kg was inactive. The results with levetiracetam substantiate similar findings of its anxiolytic actions against chlordiazepoxide withdrawal-induced anxiety in mice and in a modified Vogel test in rats and support a potential clinical use of this drug in anxiety states.

Keywords: Anxiety; Plus-maze test; Levetiracetam; Anti-epileptic; Chlordiazepoxide; Buspirone

1. Introduction

Anxiety disorder comprises a category of diverse pathological conditions in which excessive and debilitating anxiety presents as the primary symptom. Anxiety is also prominent in other psychiatric disorders and commonly occurs in various medical or neurological conditions. Pharmacological therapy for anxiety disorders is dominated by the use of benzodiazepines, buspirone and antidepressant drugs, with the newer selective serotonin re-uptake inhibitors currently in favour (Argyropoulos et al., 2000; Gelenberg, 2000). The recognition that currently available drugs are less than ideal prompts continued efforts to research improved treatments. A recent survey (Bandelow et al., 2002) refers to the anxiolytic efficacy of anticonvulsant drugs, including carbamazepine, valproate, lamotrigine and gabapentin, in preliminary studies and recommends further research.

Levetiracetam (Keppra®), the *S*-enantiomer of α-ethyl-2-oxo-1-pyrrolidine acetamide, belongs to the new generation of anti-epileptic drugs and has been approved for use as adjunctive therapy in adult patients with partial onset seizures (Dooley and Plosker, 2000). Levetiracetam is reported to be well tolerated and to significantly improve the quality of life, including reducing seizure worry (Cramer et al., 2000).

Consistent with incidental clinical data indicating potential anxiolytic activity, a recent paper (Lamberty et al., 2002) reported positive effects of levetiracetam on chlordiazepoxide withdrawal-induced anxiety assessed in mice using the elevated plus-maze. The plus-maze is probably the most widely used animal test of potential anxiolytic activity and was developed in the 1980s (Handley and Mithani, 1984; Pellow et al., 1985), having been inspired by original work by Montgomery and Monkman (1955). The test is based on exploration by rats or mice of a cross-shaped maze in which two opposite arms are enclosed while the other two are open. The maze is typically elevated leaving the animal exposed and potentially vulnerable when it ventures onto the open arms. Pharmacological validation has demonstrated that anxiolytic drugs, principally benzodiazepines, increase exploration of the open arms, whereas anxiogenic treatments have the converse effect. The ability of the plus-maze test to detect potential anxiolytic activity of anticon-

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doi:10.1016/j.ejphar.2003.09.004

vulsant drugs has been reported for carbamazepine (Zangrossi et al., 1992), gabapentin (Singh et al., 1996), tiagabine (Schmitt et al., 2002), valproate (Dalvi and Rodgers, 2001) and vigabatrin (Sayin et al., 1992).

The clinical relevance of the anxiety engendered in the plus-maze is the subject of debate (Dawson and Tricklebank, 1995) and has provoked fundamental research to determine the nature of the response and the conditions, which influence the behaviour and drug effects. One criticism is that the test is measuring the normal response of a non-pathological animal faced with a threatening situation, and therefore, drugs which affect such responding are, in effect, interfering with normal adaptive processes. In support of this, we have previously reported resistance of baseline activity to exogenous influences, such as changes in light intensity, height of maze or pre-exposure to an acute stressful treatment such as partial immersion in water or immobilisation (Falter et al., 1992). In contrast, as we now report in the present paper, the aversive nature of the open arms can be enhanced by previous exposure of rats to a four-open-arm form of the plus-maze at least 24 h prior to testing on the standard plus-maze. Under these conditions of enhanced anxiety, a clear anxiolytic profile of levetiracetam could be demonstrated.

2. Methods and materials

2.1. Animals

Young male SPF Sprague–Dawley rats (OFA origin, supplied by IFFA CREDO, Belgium) were used. At the beginning of the experiments, the rats were 6 to 8 weeks old and weighed approximately 200 ± 20 g. During the week preceding the experiment, they were housed in groups of up to eight rats per cage in wire mesh cages (dimensions $40 \times 40 \times 20$ cm high). The animals were allowed ad lib access to food and water and maintained under a 12-h light–dark cycle with lights on at 06:00 h.

2.2. Drugs and solutions

Levetiracetam (UCB Pharma), chlordiazepoxide hydrochloride (RBI) and buspirone hydrochloride (Bristol-Myers) were all dissolved in 0.9% saline solution. A dose-volume of 5 ml/kg body weight was used. Control groups received an equivalent dose-volume of 0.9% saline solution. Drugs were administered by intraperitoneal injection (i.p.). The doses of levetiracetam chosen were those reported to be active in various animal models of epilepsy (Gower et al., 1992; Klitgaard et al., 1998).

2.3. Apparatus

An elevated plus-maze made of plastic was used which consisted of four arms each 15 cm wide and 45 cm long, positioned to form a square cross around a 15-cm centre square. The plus-maze was mounted 70 cm above the floor. The maze was illuminated by uniform lighting of low intensity (9 to 12 lx). Activity on the plus-maze was observed by means of a camera which relayed via a monitor to the observer located in the adjacent room. The standard form consisted of two open arms and two closed arms, with like arms opposite one another. The closed arms were equipped with 15-cm-high walls covered with black paper. The floor of the maze was covered with black rubber. In some experiments, the standard maze was transformed either into a four-open-arm maze by replacing the closed arms with open arms or vice versa, into a four-closed-arm maze by replacing the open arms with closed arms. To avoid possible inter-observer differences, the same observer was used throughout the experiments described below (reliability coefficient: $r_p = 0.97$, $n = 10$).

2.4. Procedure

The effects of levetiracetam were first investigated using the standard plus-maze procedure. Thus, animals were treated with either vehicle control, levetiracetam at 17 and 54 mg/kg or chlordiazepoxide at 5 mg/kg. One hour later, each rat was subjected to a 4-min test session on the standard two-open/two-closed-arm plus-maze. This consisted of placing the animal on the centre square facing a closed arm and recording the number of entries on to the open arms and the number of entries on to the closed arms and the total time spent on the open arms. An entry was scored when a rat had entered the arm with all four feet. Under these conditions, as illustrated by this experiment, levetiracetam had no significant effect although the ability of chlordiazepoxide to increase open-arm exploration was confirmed.

We therefore explored ways of increasing the baseline anxiety level to reduce open-arm exploration. With this objective, two experiments were carried out to determine the influence of pre-exposure of the rats to various conditions prior to testing on the standard plus-maze. In the first experiment, rats were either left undisturbed in their home cage, placed for 4 min in a clean transit cage placed in the experimental room or allowed 4-min exploration on the standard plus-maze. Since this experiment showed that pre-exposure to the plus-maze significantly reduced activity on the open arms, we then investigated the relative importance of exposure to the open versus the closed arms of the maze. The second experiment therefore involved comparing the effects of exposure of rats for 4 min to either a four-open-arm maze or a four-closed-arm maze. In each experiment, 24 h after the preliminary manipulations, the rats were subjected to a 4-min session on the standard two-open-arm/two-closed-arm plus-maze as described above.

Since these two experiments showed that pre-exposure to the open arms was instrumental in increasing the baseline anxiety in terms of reducing exploration of the open arms on the standard plus-maze, we then re-investigated the effects

Table 1
Effect of levetiracetam administered 60 min prior to testing on standard plus-maze

Treatment mg/kg	%Visits on open arms	Time spent on open arms (s)	Total visits on closed arms
Control	24.3 (3.4)	17.7 (4.6)	6.8 (0.8)
Levetiracetam 17	24.5 (2.7)	19.1 (4.4)	7.4 (0.8)
Levetiracetam 54	23.4 (3.6)	18.0 (4.3)	8.4 (0.9)
Chlordiazepoxide 5	43.7 (4.3)[a]	41.5 (6.5)[a]	7.4 (0.7)

Results are means for groups of 13 naive rats with S.E.M. in parentheses.
[a] $P<0.01$ compared to Control; Student's *t*-test.

of levetiracetam in rats which had been pre-exposed to a four-open-arm maze. Thus, each rat was first subjected to a pre-exposure session consisting of 4 min on the four-open-arm maze. Twenty four hours later, the animals were given drug treatment comprising levetiracetam 5.4, 17 or 54 mg/kg or chlordiazepoxide 5 mg/kg, followed after 60 min by a 4-min session on the standard plus-maze. During the standard plus-maze session, the number of open-arm entries, total arm entries and total time spent on the open arms were recorded. The effects of buspirone 0.1, 0.5 and 1.0 mg/kg were likewise investigated in rats pre-exposed to a four-open-arm maze.

The effects of levetiracetam on standard plus-maze exploration were also determined 7 days after pre-exposure to the four-open-arm maze to investigate the durability of the pre-exposure effect. Additional control experiments were conducted consisting of injecting levetiracetam and chlordiazepoxide 60 min prior to, or immediately after, a 4-min pre-exposure session on the four-open-arm maze with testing on the standard plus-maze conducted 24 h later.

2.5. Statistical analysis

For each rat, the percentage of visits (entries) to the open arms was calculated (number of visits to open arms divided by total number of visits × 100). For each treatment group, the mean and S.E.M. was calculated for the three dependent variables. The data were statistically analysed by a one-way analysis of variance (ANOVA) with post hoc Dunnett's test performed in case of an overall significant effect of ANOVA. In instances where only two groups were compared, Student's *t*-test was applied. Student's *t*-test was also used in experiments in which the chlordiazepoxide-treated group served as the positive control and was compared directly with vehicle control.

Table 2
Effect of pre-exposure of rats to various conditions 24 h prior to testing on standard plus-maze

Group	%Visits on open arms	Time spent on open arms (s)	Total visits on closed arms
Experiment 1			
Home cage	16.9 (2.8)	13.2 (2.9)	7.9 (0.6)
Transit cage	17.0 (2.2)	14.3 (2.0)	8.9 (0.6)
Standard plus-maze	0.1 (0.1)[b]	0.2 (0.2)[b]	4.3 (0.9)[b]
Experiment 2			
Four-closed-arm maze	18.9 (3.2)	10.8 (2.2)	6.6 (0.7)
Four-open-arm maze	9.6 (3.3)[a]	4.1 (1.5)[b]	6.6 (0.8)

Results are means for groups of 12 rats with S.E.M. in parentheses.
Experiment 1: [b] $P<0.01$ compared to home cage; Dunnett' test.
Experiment 2: [a] $P<0.05$; [b] $P<0.01$ compared to four-closed-arm maze; Student's *t*-test.

3. Results

Administration of levetiracetam at 17 and 54 mg/kg had no effect (ANOVA, $P>0.05$) on plus-maze activity under

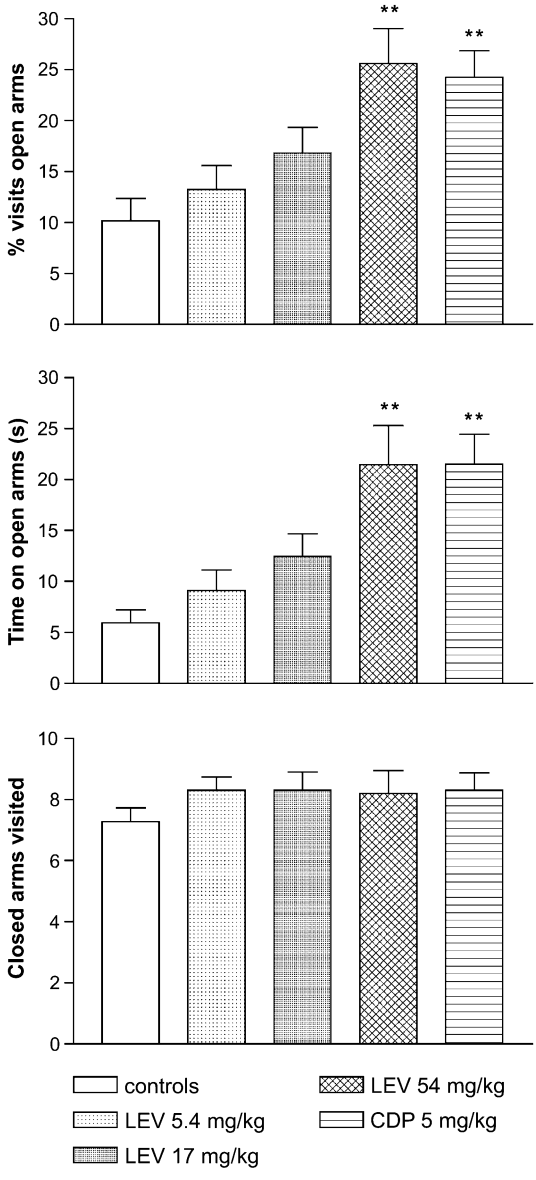

Fig. 1. Effect of levetiracetam (LEV) 5.4 to 54 mg/kg and chlordiazepoxide (CDP) 5 mg/kg on exploration of the standard plus-maze. The rats were exposed 24 h earlier to a four-open-arm maze for 4 min. Results are expressed as mean ± S.E.M. $^{**}P<0.01$ (Dunnett or Student's *t*-test) compared with saline control group.

standard conditions in which naive rats were treated acutely and then tested on the plus-maze (Table 1). Chlordiazepoxide 5 mg/kg, used as a positive control, significantly increased exploration of the open arms, both in terms of %visits to the open arms (Student's *t*-test, one-tailed, $t=3.57$, $P<0.01$) and the time spent on the open arms ($t=2.99$, $P<0.01$).

As shown in Table 2, Experiment 1, pre-exposure of rats to the plus-maze 24 h before testing on the standard plus-maze decreased exploration of the open arms (%open-arm visits: $F[2,33]=21.9$, $P<0.001$, Dunnett's test, $P<0.01$; time spent on open arms: $F[2,33]=14.7$, $P<0.001$, Dunnett's test, $P<0.01$) but also significantly decreased the number of visits to the closed arms ($F[2,33]=11.8$, $P<0.001$, Dunnett's test, $P<0.01$). A comparison of pre-exposure to a four-closed-arm maze or a four-open-arm maze showed that when tested 24 h later on the standard plus-maze, open-arm exploration was significantly lower in those rats which had been pre-exposed to the four-open-arm maze (%open-arm visits: $t=2.02$, $P<0.05$; number of open-arm visits: $t=2.66$, $P<0.01$). However, there was no significant difference in the number of closed arm visits (Student's *t*-test, $P>0.05$).

In rats exposed 24 h earlier to a four-open-arm maze, treatment with levetiracetam 5.4 to 54 mg/kg (Fig. 1) resulted in dose-dependent increases in exploration of the open arms when tested on the standard plus-maze (%open-arm visits: $F[3,101]=5.2$, $P<0.01$; time spent on open arms: $F[3,101]=7.1$, $P<0.001$), reaching statistical significance at 54 mg/kg (%open-arm visits: Dunnett's test, $P<0.01$; time spent on open-arm visits: Dunnett's test, $P<0.01$). The number of visits to the closed arms was unchanged (ANOVA, $P>0.05$). Similar significant increases in open-arm exploration occurred in rats treated with chlordiazepoxide 5 mg/kg (%visits: $t=4.2$, $P<0.01$; time spent on open arms: $t=4.8$, $P<0.01$). Under similar testing conditions, buspirone 0.1, 0.5 or 1.0 mg/kg (Table 3) had no statistically significant effect (ANOVA, $P>0.05$) although positive effects of chlordiazepoxide 5 mg/kg confirmed the validity of the experiment.

In the experiment in which rats were exposed to a four-open-arm maze 7 days prior to evaluation of effects of treatment on standard plus-maze exploration, exploration by control rats of the open arms remained low, showing the durability of the pre-exposure effect (Fig. 2). Furthermore, levetiracetam dose-dependently increased open-arm exploration reaching statistical significance at 54 mg/kg (%open-arm visits: $F[2,42]=3.9$, $P<0.05$, Dunnett's test, $P<0.05$; time spent on open arms: $F[2,42]=4.6$, $P<0.01$, Dunnett's test, $P<0.01$), thereby confirming the anxiolytic effects of levetiracetam.

Additional experiments showed that neither treatment with levetiracetam or chlordizepoxide, prior to, or immediately after, exposure on the four-open-arm maze had any effect on standard plus-maze exploration when tested 24 h later (data not shown).

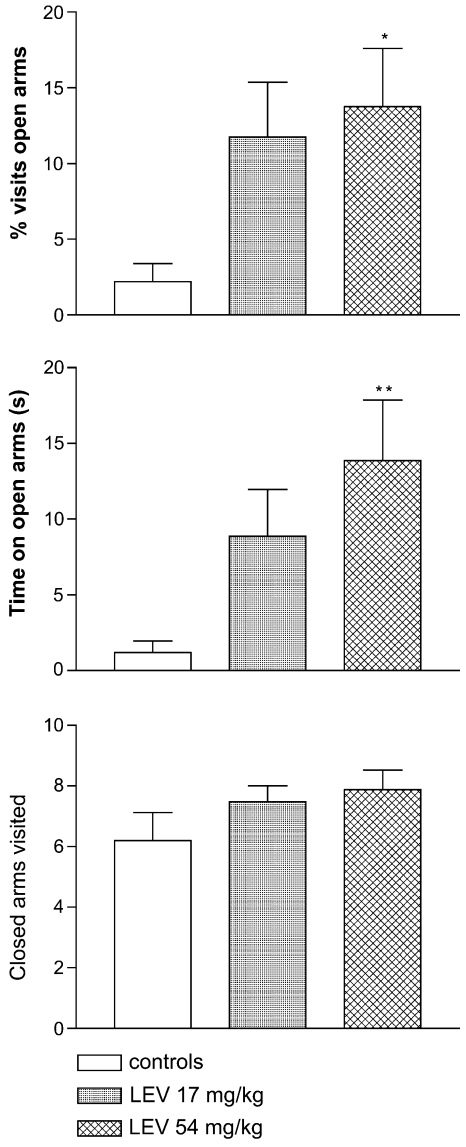

Fig. 2. Effect of levetiracetam (LEV) 17 and 54 mg/kg on exploration of the standard plus-maze. The rats were exposed 7 days earlier to a four-open-arm maze for 4 min. Results are expressed as mean ± S.E.M. *$P<0.05$; **$P<0.01$ (Dunnett's test) compared with saline control group.

Table 3
Effect of buspirone administered 60 min prior to standard plus-maze testing in rats exposed to four-open-arm maze 24 h prior to treatment

Treatment (mg/kg)	%Visits on open arms	Time spent on open arms (s)	Total visits on closed arms
Control	8.7 (3.5)	5.7 (2.4)	6.7 (0.7)
Buspirone 0.1	4.9 (2.2)	2.8 (1.5)	7.4 (0.6)
Buspirone 0.5	4.7 (2.5)	5.4 (3.7)	6.0 (0.8)
Buspirone 1.0	7.6 (2.9)	8.5 (4.2)	6.8 (0.7)
Chlordiazepoxide 5	17.0 (2.4)[a]	18.5 (3.5)[b]	9.7 (1.1)[a]

Results are means for groups of 15 rats with S.E.M. in parentheses.
[a] $P<0.05$ compared to Control; Student's *t*-test.
[b] $P<0.01$ compared to Control; Student's *t*-test.

4. Discussion

The results of the present study revealed an anxiolytic action of levetiracetam in the standard plus-maze which is contingent upon the experimental conditions. Thus, levetiracetam increased open-arm exploration in rats in which the baseline had been lowered by their prior exposure to a four-open-arm maze. In contrast, the routine approach of using rats naive to the plus-maze proved resistant to the effects of levetiracetam. Although the present paper reports an experiment in which levetiracetam was inactive, in practice, it is possible to observe an anxiolytic effect of levetiracetam under standard conditions. Notwithstanding, the effect is difficult to reproduce and is most likely to be observed when the baseline exploration of control animals is low (personal observations; data not shown).

This observation led us to conduct a series of experiments to establish the conditions under which the baseline is consistently lowered. This was achieved by exposure of the rats to a four-open-arm form of the maze 24 h before drug treatment and testing. Under these conditions of enhanced anxiety, levetiracetam dose-dependently increased open-arm exploration, thereby supporting an anxiolytic action. Importantly, levetiracetam treatment did not affect locomotor activity, assessed in terms of visits to the closed arms. Supported by factor analysis (Rodgers and Johnson, 1995), closed arm activity is considered a superior measure of locomotor effects than total arm visits since this latter measure includes the open arms which are subject to anxiolytic and anxiogenic influences. Chlordiazepoxide had similar positive anxiolytic effects. The effects of levetiracetam were replicated when the exposure-test interval was extended to 7 days, showing that this is a robust effect. For the record, subsequent experiments (data not shown) using open-arm pre-exposure are able to reproduce the anxiolytic effects of levetiracetam in the plus-maze and substantiate the initial observation that reproducibility depends on an enhanced baseline level of anxiety.

The absence of effect of either levetiracetam or chlordiazepeoxide when given either before or immediately after exposure to the four-open-arm maze indicates that the positive effect is not attributable to treatment-induced changes in the experience of prior exposure, such as amnesia.

Unlike levetiracetam or chlordiazepoxide, buspirone did not present anxiolytic effects when tested in rats previously exposed to the four-open-arm maze. Similar negative results were obtained when buspirone was tested in naive rats (data not shown). Although an accepted clinical treatment for generalised anxiety (Bandelow et al., 2002), buspirone produces variable effects in animal tests for anxiolytic activity. For example, in the plus-maze, effects of buspirone have ranged from anxiolytic through to anxiogenic (Critchley and Handley, 1987; Dunn et al., 1989; Moser, 1989; Moser et al., 1990; Pellow and File, 1986). A recent paper by Haller et al. (2000) indicated that anxiolytic effects of buspirone in the plus-maze could be demonstrated in isolated but not group-housed rats. The expectation of increased emotionality in isolated rats could predict positive anxiolytic effects of buspirone in the present study using pre-exposed rats, which was not the case. However, since baseline plus-maze exploration was not significantly altered by isolation, the authors concluded that increased anxiety levels did not account for increased efficacy of buspirone in isolated rats. The doses of 3 and 10 mg/kg of buspirone used by Haller et al. (2000) are higher than those of 0.1 to 1.0 m/kg used in the present study. Nevertheless, the inability to demonstrate an anxiolytic effect of buspirone in unlikely to be due to dose since the doses used are ones which have previously been shown to be anxiolytic in the plus-maze under normal conditions (Kostowski et al., 1992) or under ethanol-induced withdrawal (Lal et al., 1991).

The interesting pharmacological profile of levetiracetam was first reported in 1992 (Gower et al., 1992) and has since been extensively investigated in in vivo and in vitro models of convulsant activity (for review, see Klitgaard, 2001; Margineanu and Klitgaard, 2002). Although a distinct central binding site for levetiracetam has been identified (Noyer et al., 1995), its mechanism of action has yet to be established but appears to exclude those of other anti-epileptic drugs to involve actions such as direct activation of γ-aminobutyric acidergic (GABAergic) systems or inhibition of excitatory mechanisms (Margineanu and Klitgaard, 2002). Notwithstanding, some involvement with GABA has been implicated; for example, levetiracetam induced alterations in GABA metabolism and turnover, albeit at relatively high doses (Loscher et al., 1996). Recent work (Rigo et al., 2002) implicates an ability of levetiracetam to reverse the inhibitory effects of negative allosteric modulators of neuronal GABA- and glycine-gated channels. GABAergic systems are implicated not only in epilepsy but also in anxiety disorders, exemplified through benzodiazepines acting on the benzodiazepine–GABA–Cl^- complex. In this context, it is interesting to note that the anticonvulsant drugs, gabapentin, tiagabine and vigabatrin which may act via GABAergic mechanisms (see Mehta and Ticku, 1999), were all effective on anxiety generated by the plus-maze test (Sayin et al., 1992; Schmitt et al., 2002; Singh et al., 1996).

A lot of research has been directed at investigating the nature of anxiety engendered by the plus-maze, although the data and conclusions are frequently conflicting. Our own investigations (Falter et al., 1992) showing lack of effect of various exogenous factors on plus-maze activity led us to the conclusion that plus-maze anxiety was a situational-bound response. The present results of reduced open-arm exploration after exposure to a four-open-arm maze are not incompatible with that conclusion although clearly pre-exposure resulted in an enhanced anxiety state. The difference between the present and previous findings is explicable in terms of the nature of the stressor to which the animals were exposed prior to standard plus-maze testing. In a recent

review, Korte and De Boer (2003) showed that the nature of the stressor is an important factor in determining fear potentiation in the plus-maze. These authors reported that robust fear potentiated behaviour in the plus-maze is produced in rats previously subjected to inescapable footshock but is absent in rats pre-exposed to controllable footshock. To some extent, pre-exposure to the open-arm maze, in which the animal is unable to avoid the aversive open-arms, is analogous to inescapable footshock.

There are several reports which are at variance with the present finding of enhanced anxiety induced by pre-exposure. For example, Pellow et al. (1985) and later File et al. (1992) from the same laboratory reported no change in the baseline activity after repeated exposure, a finding which the authors themselves recognised as surprising as some habituation would be expected. Lister (1987) reported a similar absence of pre-exposure effect in mice. In direct contrast, however, more recent reports concur with the present study that prior plus-maze experience causes significant reductions in open-arm exploration in both rats (Shepherd, 1992; Griebel et al., 1993; Treit et al., 1993; Bertaglio and Carobrez, 2000; Dal-Cól et al., 2003) and mice (Rodgers and Shepherd, 1993; Rodgers et al., 1997; Holmes and Rodgers, 2003). It is difficult to identify any particular factor which would account for the variability in results. As discussed by Griebel et al. (1993), there are several factors which could affect either or both baseline and potential drug effects.

In addition to baseline changes, previous exposure to the plus-maze has been reported to lower or even eliminate the anxiolytic efficacy of benzodiazepines in rats (File et al., 1992; File and Zangrossi, 1993) or mice (Rodgers and Shepherd, 1993). The present data suggest a reduced effect of chlordiazepoxide in pre-exposed rats although a statistically significant anxiolytic effect remained evident. In view of recognised variability within this test, it is not possible to say whether this is a real decrease. Dawson and Tricklebank (1995) concluded that an anxiolytic effect of diazepam in pre-exposed rats is still observable in the study by File et al. (1993) if changes in locomotor activity are taken into account. One major difference in the testing conditions was that in the present study, drug effects were investigated in rats pre-exposed to a four-open-arm maze, rather than the standard plus-maze. This could represent an advantage in reducing the impact of habituation as a confounding factor while increasing the fear potentiating impact of the open-arms in subsequent testing. In this context, it is pertinent to note that Bertaglio and Carobrez (2002) recently reported that loss of anxiolytic effects of the benzodiazepine, midazelam, occurred in rats previously exposed to the standard plus-maze, whereas its anxiolytic effects were retained in rats previously exposed to either open or closed arms. This finding is consistent with the present study showing anxiolytic effects of chlordiazepoxide in rats pre-exposed to four-open-arm maze. The present results of increased sensitivity to drug effects in maze-experienced animals are not unique. For example, Rodgers et al. (2003) recently reported that a single undrugged experience of the elevated plus-maze allowed anxiolytic effects of a cannabinoid receptor antagonist to be revealed. Furthermore, the same experimental pre-exposure conditions strengthened anxiogenic effects of phenelzine and fluoxetine (Holmes and Rodgers, 2003).

Taking account of both behavioural and neurochemical changes, File et al. (1993) proposed that the fear produced on re-testing in the plus-maze is qualitatively different from that produced in naive rats and suggest that trial 2 anxiety might be equivalent to phobic anxiety. The arguments for this are the reduction of efficacy of benzodiazepines in re-tested animals and their relative lack of efficacy against phobias in the clinic. The data reviewed by Korte and De Boer (2003) likewise differentiate between anxiety measured by the traditional (spontaneous) elevated plus-maze and the enhanced anxiety state of fear potentiated behaviour.

Extrapolating from this, levetiracetam would be expected to be particularly effective against anticipatory anxiety. Such an expectation is supported by other rodent studies showing an anxiolytic effect of levetiracetam. Thus, levetiracetam counteracted chlordiazepoxide withdrawal-induced anxiety assessed in the plus-maze using mice but had no effect on "normal" mice (Lamberty et al., 2002). Similarly, an anxiolytic effect in the Vogel conflict test using rats was contingent upon increased anxiety produced by administration of an electric shock at the end of the habituation period prior to testing (Lamberty et al., 2003). The experimental conditions required to demonstrate an anxiolytic action of levetiracetam all support a dependence on enhanced emotionality.

These data therefore provide evidence for a potential clinically relevant anxiolytic action of levetiracetam which merits further investigation. In addition, the contingency of these anxiolytic effects on the experimental conditions of testing is particularly pertinent to an experimental paradigm which, while apparently simple to perform, has produced such markedly varying pharmacological results.

Acknowledgements

We gratefully acknowledge the technical assistance of Anne-Catherine Michaux and François Druant.

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