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Piracetam and levetiracetam, two pyrrolidone derivatives, exert antidystonic activity in a hamster model of paroxysmal dystonia

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Abstract

The effects of the nootropic drug piracetam and its analogue, the antiepileptic drug levetiracetam (ucb L059) on severity of dystonic attacks were studied in a mutant hamster model of idiopathic generalized dystonia. Both drugs significantly decreased the severity of dystonia. In contrast to seizure models, in which levetiracetam is much more potent as an anticonvulsant than piracetam, the antidystonic potency of levetiracetam was only moderately higher than that of piracetam. The antidystonic activity of piracetam and levetiracetam was not associated with any behavioral side effects. The data indicate that piracetam and levetiracetam are interesting novel treatments for idiopathic dystonia.

Keywords: Movement disorder; Nootropic drug; Epilepsy; Anticonvulsant drug

1. Introduction

Dystonia is a common movement disorder, characterized by sustained muscle contractions, frequently causing twisting and repetitive movements or abnormal postures (Fahn et al., 1987). About two-third of cases are idiopathic, i.e., occur in the absence of lesions within the central nervous system (McGeer and McGeer, 1995). The anatomical and neurochemical basis for idiopathic dystonia is unknown (McGeer and McGeer, 1995). Due to the lack of knowledge about the pathophysiology of idiopathic dystonia, rational therapies are not available, and the empirical treatment of idiopathic dystonia, particularly generalized dystonia, is often disappointing (McGeer and McGeer, 1988). Animal models of dystonia may contribute to insights into the pathophysiology of dystonia and can be helpful in the development of new therapeutic strategies (Richter and Löscher, 1998).

We have previously characterized a genetic hamster model of paroxysmal dystonia (paroxysmal dystonic choreoathetosis), a form of idiopathic generalized dystonia (Richter and Löscher, 1998). Drugs that are used for treatment of this disorder in humans are also effective in the hamster model, suggesting that this model may be used for testing of new therapeutic agents (Richter and Löscher, 1998). In the present study, we tested the novel anticonvulsant drug levetiracetam (ucb L059), an analogue of the nootropic (cognition-enhancing) agent piracetam (Haria and Balfour, 1997) in mutant dystonic hamsters. This was prompted by a previous anecdotal report on piracetam for treatment of choreoathetosis (Piperidou et al., 1988) and the fact that levetiracetam is much more potent than piracetam in animal models of epilepsy or seizures (Löscher and Hönack, 1993; Gower et al., 1995). In order to test whether levetiracetam is also a more effective antidystonic agent than piracetam, the latter drug was included in the present experiments for comparison.

2. Materials and methods

2.1. Animals

The mutant dt^{sz} hamsters used in the present experiments were obtained by selective breeding as described in detail previously (Fredow and Löscher, 1991). All hamsters were housed in groups of three to five animals in

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Table 1
Effect of piracetam or levetiracetam on the latency to onset of dystonic attacks in mutant dystonic hamsters. Latency was determined as the time to the first unequivocal signs of the dystonic attack (stage 2). Control latencies were recorded 2 days before each drug trial. Dystonic attacks were induced by a triple-stimulation procedure (see Methods). Data are means ± S.E. of 10–14 hamsters per experiment

Drug	Dose (mg/kg i.p.)	Latency (min)		Significance
		Pre-drug trial	Drug trial	
Piracetam	54	10.2 ± 2.1	16.1 ± 4.6	n.s.
	108	12.6 ± 1.6	22.3 ± 2.6	$P = 0.0098$
Levetiracetam	13.5	22.9 ± 3.9	16.3 ± 1.9	n.s.
	27	20.3 ± 0.7	27.8 ± 5.6	n.s.
	54	12.3 ± 1.7	12.4 ± 1.6	n.s.
	108	8.8 ± 1.7	31.5 ± 9.9	$P = 0.0023$

plastic cages at an ambient temperature of 23–25°C with a light cycle of 13 h (light on at 6:00 a.m.), and were fed on Altromin 1320 standard diet (Altromin, Lage, Germany). All behavioral observations were carried out in the forenoon between 9 and 12 a.m. at a controlled temperature (23–25°C). In all experiments, animal groups consisted of male and female hamsters, since there was no indication of sex-related differences in dystonia or the effect of drugs on dystonia. All animal care and handling was conducted in compliance with the German Animal Welfare Act and was approved by the responsible governmental agency in Hannover.

2.2. Drug testing in dystonic hamsters

Drug testing was performed during the age of maximal sensitivity of the hamsters for induction of dystonic attacks, i.e., between 30 and 40 days of age (cf. Richter and Löscher, 1998). Groups of 10–14 dystonic hamsters were used per experiment. For induction of dystonic attacks, the hamsters were taken from their home cage and placed on a balance (for determining body weight), were then injected i.p. with saline (controls) or drug, and immediately placed in a new (clean) and empty plastic cage (one animal per cage). As previously described (Fredow and Löscher, 1991), this ''triple stimulation'' procedure resulted in the induction of dystonic attacks within minutes after placing the animals in the new cage. The animals were observed in this cage for 3 h, and the severity of the dystonic movements was rated as follows: stage 1, flattened ears and flattened posture while walking; stage 2, facial contortions, rearings with forelimbs crossing, disturbed gait with retarded setting of forepaws; stage 3, stiffened hindlimbs so that the animals appear to walk on tiptoes in a dysmetric hypergait; stage 4, loss of balance; stage 5, hindlimbs hyperextended caudally, animal continues to pull itself with the functional forelimbs; stage 6, animal immobilized in a twisted, hunched posture with both hindlimbs and forelimbs tonically extended forward, erected (Straub-like) tail, alternating unilateral forelimb elevation, head weaving, and opisthotonus. This final stage persisted for 2–5 h, but rapid recovery occurred thereafter. All mutant hamsters did not progress through the entire sequence described; the individual maximum stage was usually reached within 45–170 min.

Piracetam was tested at two doses, 54 and 108 mg/kg i.p., in dystonic hamsters. Levetiracetam was tested at four doses, i.e., 13.5, 27, 54, and 108 mg/kg i.p. In addition to recording of the dystonic attacks, all hamsters were closely observed for behavioral adverse effects caused by the drug treatments. The doses of levetiracetam and piracetam evaluated in this study in hamsters were based on previous experiments with these drugs in rodent seizure models (Löscher and Hönack, 1993).

2.3. Drugs

Piracetam and levetiracetam were obtained from UCB Pharma (Braine-l'Alleud, Belgium) and were freshly dissolved in distilled water prior to each injection. Injection volume was 5 ml/kg. In control trials, hamsters received the same volume of saline.

2.4. Statistics

Significance of differences between severity scores in control trials and drug trials in the same group of hamsters was calculated by the Wilcoxon rank test for paired repli-

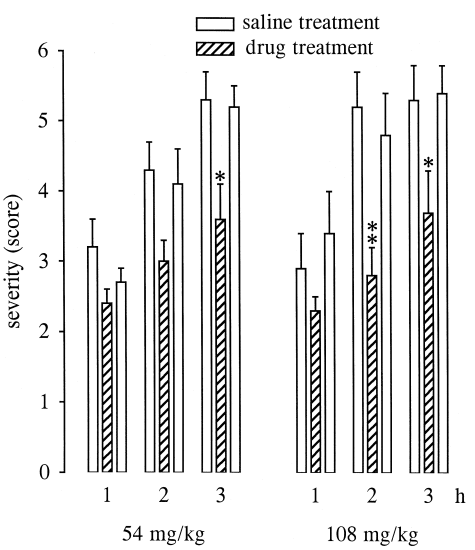

Fig. 1. Effect of piracetam on severity of dystonia in mutant hamsters at the age of maximum sensitivity for induction of dystonic attacks (30–40 days). The figure shows the average of the maximum individual severity scores of dystonia reached within the first, second and third hour(s) after i.p. administration of 54 or 108 mg/kg. Control recordings were taken 2 days before (pre-drug control) and 2 days after (post-drug control) the drug trial. Asterisks indicate significant reduction in severity in comparison to the pre-drug and post-drug control ($^{*}P < 0.05$; $^{**}P < 0.01$). Data are shown as means ± S.E. of 11 (54 mg/kg) or 10 (108 mg/kg) dystonic hamsters. Open bars: pre- and post-drug control scores; hatched bars: scores under treatment.

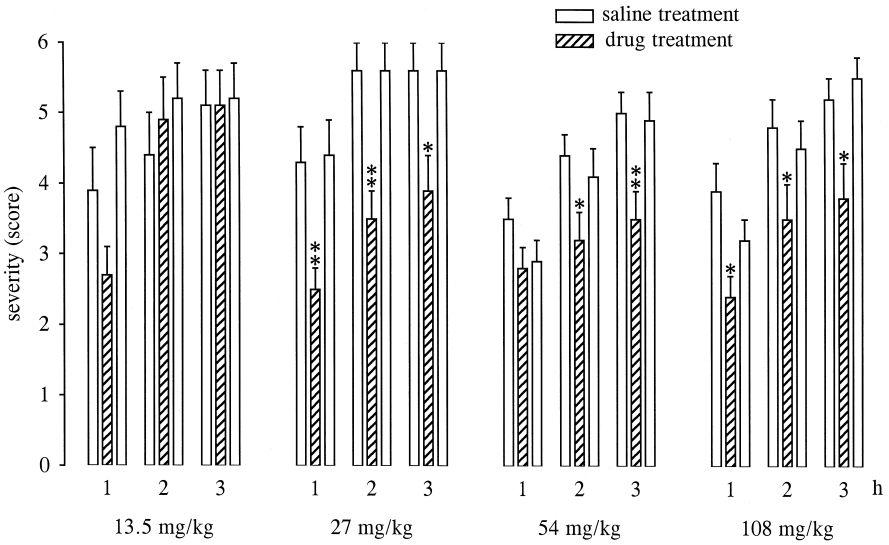

Fig. 2. Effect of levetiracetam on severity of dystonia in mutant hamsters at the age of maximum sensitivity for induction of dystonic attacks (30–40 days). The figure shows the average of the maximum individual severity scores of dystonia reached within the first, second and third hour(s) after i.p. administration of 13.5, 27, 54, or 108 mg/kg. Control recordings were taken 2 days before (pre-drug control) and 2 days after (post-drug control) the drug trial. Asterisks indicate significant reduction in severity in comparison to the pre-drug and post-drug control ($^{*}P < 0.05$; $^{**}P < 0.01$). Data are shown as means ± S.E. of 10 (13.5, 24 mg/kg), 14 (54 mg/kg) or 13 (108 mg/kg) dystonic hamsters. Open bars: pre- and post-drug control scores; hatched bars: scores under treatment.

cates. This test was also used to calculate significant differences in latencies to onset of dystonic attacks in control and drug trials. Because we assumed that the drugs would exert antidystonic effects, all tests were used one-sided and a P of at least 0.05 was considered significant.

3. Results

Piracetam did not retard the onset of dystonic attacks at 54 mg/kg, but caused a significant retardation at 108 mg/kg (Table 1). The severity of dystonic attacks was significantly reduced by both doses (Fig. 1). No behavioral adverse effects were induced by piracetam in the hamsters.

Levetiracetam significantly increased the latency to onset of dystonic attacks only at the highest dose tested (Table 1). With respect to severity of dystonic attacks, the lowest dose of levetiracetam (13.5 mg/kg) tended to reduce severity during the first hour after administration, which, however, was only statistically significant when compared to the post-drug trial (Fig. 2). The higher doses of levetiracetam, i.e., 27, 54, and 108 mg/kg, significantly decreased severity of dystonic attacks both in comparison to pre- and post-drug control trials (Fig. 2). At doses of 13.5–54 mg/kg, no behavioral adverse effects were observed. At 108 mg/kg, moderate hyperactivity and slight ataxia were seen in some of the animals.

4. Discussion

The present data on piracetam in mutant dystonic hamsters are in line with a previous clinical observation (Piperidou et al., 1988), indicating that the nootropic drug piracetam exerts antidystonic activity. Significant antidystonic effects of piracetam in mutant hamsters were determined at both doses tested, i.e., 54 and 108 mg/kg. These doses are clearly below those previously reported to exert anticonvulsant effects in rodent models of epilepsy (Schmidt, 1990; Fischer et al., 1991; Keller, 1991; Löscher and Hönack, 1993). For instance, doses of below 200 mg/kg of piracetam were ineffective in the kindling model of epilepsy (Schmidt, 1990; Löscher and Hönack, 1993). In humans, piracetam (2.4 g/day) has been reported to improve seizure protection in epileptic patients receiving carbamazepine (Chaudry et al., 1992). Doses of up to 45 g piracetam daily have been successfully used in patients with cortical myoclonus without significant adverse effects during long-term use (Genton et al., 1999). Much lower doses (2–4 g) have been anecdotally reported to exert therapeutic efficacy in human choreoathetosis (Piperidou et al., 1988). On a milligram per kilogram body weight basis, the latter doses (about 30–60 mg/kg) are in the dose range found efficacious in the present hamster study.

Levetiracetam, an analogue of piracetam, is at least 10-times more potent as an anticonvulsant than piracetam in seizure models (Löscher and Hönack, 1993; Gower et al., 1995). In clinical trials, levetiracetam proved efficacious in patients with partial epilepsy (Perucca, 1999) and juvenile myoclonus epilepsy (Smith and Betts, 1998). In contrast to the marked difference in anticonvulsant potency between levetiracetam and piracetam in seizure models, levetiracetam exhibited similar antidystonic potency and efficacy as piracetam in the present hamster experiments. This might indicate that the mechanisms involved in the

anticonvulsant actions of piracetam and levetiracetam differ from the mechanisms of their antidystonic activity. While there is some evidence that the cognition-enhancing (nootropic) efficacy of piracetam may be related to positive modulation of glutamate receptors (Copani et al., 1992) and effects on membrane fluidity (Müller et al., 1997, 1999), the mechanism of its anticonvulsant activity, for instance in treatment of cortical myoclonus, is not clear. With respect to levetiracetam, despite extensive experiments, the exact mechanism of action of this drug is unknown (Perucca, 1999). Potentiation of inhibition mediated by γ-aminobutyric acid (GABA) has been proposed (Löscher et al., 1996), but the drug had no significant affinity for known GABA, benzodiazepine, glycine, adenosine, and various excitatory amino acid-related receptors, or for Ca^{2+}, K^{+} and Cl^{-} channels (Perucca, 1999). Studies using central brain membranes have suggested that levetiracetam acts via a specific binding site in the rat brain (Noyer et al., 1995) which, however, requires further characterization. It is therefore currently not possible to judge which mechanism(s) of piracetam and levetiracetam might explain the antidystonic activity of these drugs observed in the present experiments.

The fact that the beneficial effect of piracetam in choreoathetosis observed clinically (Piperidou et al., 1988) was also observed in the dystonic hamster model substantiates the predictive character of this model and strongly suggests that levetiracetam may also be effective in dystonic patients, therefore adding a potential new indication for this interesting novel anticonvulsant agent.

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