ELSEVIER

European Journal of Pharmacology 317 (1996) 183–190

ejp

Hyperactivity and behavioral seizures in rodents following treatment with the dopamine D_1 receptor agonists A-86929 and ABT-431

Kazumi Shiosaki *, Karen E. Asin , Donald R. Britton , William J. Giardina , Lisa Bednarz , Laura Mahan , Joe Mikusa , Arthur Nikkel , Carol Wismer

Neuroscience Discovery, Pharmaceutical Products Division, D-47D, AP-9A, 100 Abbott Park, Abbott Laboratories, Abbott Park, IL 60064, USA

Received 29 August 1996; accepted 6 September 1996

Abstract

A-86929 ((−)-*trans*-9,10-dihydroxy-2-propyl-4,5,5*a*,6,7,11*b*-hexahydro-3-thia-5-azacyclopent-1-ena[*c*]phenanthrene) is a potent and selective full agonist at the dopamine D_1 receptor. Both A-86929 and ABT-431 ((−)-*trans*-9,10-diacetyloxy-2-propyl-4,5,5*a*,6,7,11*b*-hexahydro-3-thia-5-azacyclopent-1-ena[*c*]phenanthrene hydrochloride), the diacetyl prodrug derivative of A-86929, were evaluated for their effects on behavioral excitability in rodents. In rats, A-86929 produced a dose-dependent increase in locomotor activity that was attenuated by the selective dopamine D_1 receptor antagonist, SCH 23390, as well as by higher doses of the dopamine D_2 receptor antagonist, haloperidol. Repeated administration of A-86929 over 6 days produced hyperactivity which did not change in magnitude across days. Acute administration of A-86929 and ABT-431 to mice produced behavioral seizure activity, with ED_{50} values of 7.1 and 2.7 μmol/kg, s.c., respectively, that was blocked by SCH 23390. Young rats (35–37 days) exhibited behavioral seizures following A-86929 and ABT-431 treatment (ED_{50} = 34.2 and 35.6 μmol/kg, s.c., respectively), but at doses higher than those required in mice. Moreover, adult rats (3 months) were less sensitive (ED_{50} = 345 μmol/kg, s.c.) to A-86929-induced seizures than young rats. Comparison of the ED_{50} values that produced behavioral seizure activity in rats with those previously established to produce contralateral rotation (ED_{50} = 0.24 μmol/kg, s.c.) in 6-hydroxydopamine-lesioned rat indicates that a significant dose separation exists between these two properties of A-86929.

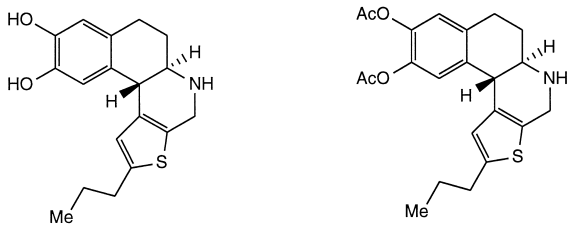
Keywords: Dopamine D_1 receptor ; A-86929 ; ABT-431 ; Hyperactivity ; Seizure; Tolerance

1. Introduction

A-86929 ((−)-*trans*-9,10-dihydroxy-2-propyl-4,5,5*a*,6,7,11*b*-hexahydro-3-thia-5-azacyclopent-1-ena[*c*]-phenanthrene) is a selective, high-affinity (K_i = 51 nM) ligand for the dopamine D_1 receptor and possesses high potency (EC_{50} = 9.0 nM) and full intrinsic activity relative to dopamine in stimulating adenylate cyclase. Both A-86929 and ABT-431 (Fig. 1), the diacetyl derivative of A-86929 that is rapidly converted to the parent compound in vivo, produced robust contralateral rotation in the 6-hydroxydopamine-lesioned rat model (ED_{50} = 0.24 and 0.54 μmol/kg, s.c., respectively), confirming the potent dopamine receptor agonist activity of the compounds in vivo (Michaelides et al., 1995; Shiosaki et al., 1996). The contralateral rotation following A-86929 or ABT-431 treatment was blocked by selective dopamine D_1, but not D_2, receptor antagonists. Additional studies with A-86929 and ABT-431 demonstrated the ability of these compounds to reverse parkinsonian-like behavioral disabilities in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-lesioned marmoset model (Shiosaki et al., 1996). Effective reversal of these MPTP-induced behavioral deficits in nonhuman primates suggests the potential usefulness of A-86929 and ABT-431 in treating Parkinson's disease (Jenner et al., 1986).

In addition to causing contralateral rotation in the 6-hydroxydopamine-lesioned rat model, dopamine D_1 receptor agonists produce behavioral activation in intact rats. Systemic administration of SKF 38393 (1-phenyl-2,3,4,5-tetrahydro-(1*H*)-3-benzazepine-7,8-diol hydrochloride), a selective dopamine D_1 receptor agonist possessing partial intrinsic activity relative to dopamine, increased grooming, sniffing and vacuous jaw movements, but generally did not

* Corresponding author. Tel.: (1-847) 937-5933; Fax: (1-847) 938-1021; e-mail: kazumi.shiosaki@abbott.com



A-86929 ABT-431

Fig. 1. Structures of A-86929 and ABT-431.

affect horizontal locomotor activity (for review see Jackson and Westlind-Danielsson, 1994). In contrast, SKF 82958 (3-allyl-6-chloro-2,3,4,5-tetrahydro-7,8-dihydroxy-1-phenyl-1*H*-3-benzazepine), which is a dopamine D_1 receptor-selective full agonist, increased locomotor activity and stereotyped behaviors following acute administration (Meyer and Shults, 1993), as did dopamine D_2 receptor-selective agonists (Eilam et al., 1992; Molloy et al., 1986).

SKF 38393 has also been reported to increase the sensitivity of rats to pilocarpine-induced behavioral seizures (Al-Tajir et al., 1990a,b; Barone et al., 1990; Turski et al., 1990) and to produce convulsions 24 h after treatment in reserpinized mice (Al-Tajir et al., 1990a,b). These effects were reversed by the dopamine D_1 receptor-selective antagonist SCH 23390. In contrast, SKF 38393 protected mice from seizures induced by pentylenetetrazole in a dose-dependent manner (Ogren and Pakh, 1993), suggesting that the involvement of dopamine D_1 receptors in the propagation or the attenuation of behavioral seizures is dependent on the specific seizure model used (Starr, 1996). SKF 82958 and the isochroman A-68930 ((1*R*,3*S*)-1-aminomethyl-3,4-dihydro-5,6-dihydroxy-3-phenyl-1*H*-2-benzopyran), produced clonic-tonic seizures when administered to neurologically intact rats and mice (Britton et al., 1991). The convulsant properties appear to be associated with dopamine D_1 receptor agonists as a class based on the ability of structurally dissimilar D_1 compounds to produce similar behavioral responses.

In the present paper, we describe the effects of A-86929 on the locomotor activity of rats following both acute and repeated administration, and on the production of behavioral seizure activity following administration of A-86929 and/or ABT-431 to mice and rats. The ability of SCH 23390 to reverse these behavioral effects was also assessed.

2. Materials and methods

2.1. Subjects

Male CD-1 mice (Charles River, Portage, MI, USA) weighing approximately 13–22 g were housed 14 per cage, with food and water available ad libitum.

Male Sprague-Dawley rats (Sasco, Madison, WI, USA) weighing 175–330 g were used in the locomotor activity studies. Young Sprague-Dawley male rats (Sasco), approximately 35–37 days old and weighing about 100 g, and adult Sprague-Dawley male rats (Sasco), approximately 3 months old and weighing about 350 g, were used in the seizure studies. The rats were group housed, approximately five per cage, with food and water available ad libitum. A 12:12 light:dark cycle (lights on at 06:00) was maintained and testing was conducted during the light portion of the cycle.

2.2. Compounds

A-86929 ((−)-*trans*-9,10-dihydroxy-2-propyl-4,5,5*a*,6,7,11*b*-hexahydro-3-thia-5-azacyclopent-1-ena[*c*]-phenanthrene) and ABT-431 ((−)-*trans*-9,10-diacetyloxy-2-propyl-4,5,5*a*,6,7,11*b*-hexahydro-3-thia-5-azacyclopent-1-ena[*c*]phenanthrene hydrochloride) were synthesized at Abbott Laboratories (Abbott Park, IL, USA) (Michaelides et al., 1995). SCH 23390 was purchased from Research Biochemicals International (Natick, MA, USA). Haloperidol was purchased from Sigma (St. Louis, MO, USA). The vehicle for ABT-431 in the seizure studies was 10% ethanol. Haloperidol was dissolved in a small quantity of 0.1 M acetic acid and then brought up to volume with sterile water. The vehicle for all other studies was sterile distilled water. Haloperidol was administered in a volume of 2 ml/kg, as was A-86929 (100 μmol/kg) in the seizure antagonist study. All other injection volumes were 1 ml/kg. All injections were placed in the subcutaneous, intrascapular space.

2.3. Locomotor activity in rats

2.3.1. Acute dose-response study

Rats were placed in testing chambers housed within ventilated, sound-attenuating isolation chambers (E10-23, Coulbourn Instruments, Allentown, PA, USA). Each chamber measured $48 \times 23 \times 25$ cm and was equipped with a photobeam that transects its length. Interruptions of the photobeam when the animal crossed over the center of the cage were recorded using an on-line monitoring system.

For the dose-response determination, rats were given a 40-min habituation period, injected with vehicle or various doses of A-86929 (0.5, 1.0, 2.0, 4.0 or 8.0 μmol/kg, s.c.), and were returned immediately to the testing chambers. Horizontal locomotor activity (crossovers) was subsequently monitored for 6 h. Eight rats were used in each treatment group.

2.3.2. Antagonism of A-86929-induced hyperactivity

To determine the effects of dopamine-receptor antagonists, rats were injected with either vehicle or various doses of SCH 23390 (0.001, 0.003, 0.01, or 0.10 μmol/kg, s.c.) after the 40-min habituation period and then returned

to the activity cage. 10 min later, animals were injected with A-86929 (1.0 μmol/kg, s.c.), returned to the activity cage and horizontal locomotor activity (crossovers) was measured for 4 h.

A second group of rats was injected with vehicle or various doses of haloperidol (0.01, 0.03, 0.10, or 1.0 μmol/kg, s.c.) following the 40-min habituation period and was then returned to the activity cages. 30 min later, animals were injected with A-86929 (1.0 μmol/kg, s.c.), returned to the activity cages and horizontal locomotor activity (crossovers) was monitored for 4 h. Each treatment group consisted of 10–12 rats.

2.3.3. Effects of subchronic A-86929 treatment on locomotor activity

In a separate study, rats were injected with vehicle or A-86929 (1.0, 3.0, or 6.0 μmol/kg, s.c.) daily for 6 days and were tested for locomotor activity across a 3 h, post-injection period on days 1, 3, and 6 as described above. Each treatment group contained 8–15 rats.

2.4. Behavioral seizure activity in mice

2.4.1. Acute dose response

Mice were injected subcutaneously with vehicle or various doses of A-86929 (0.30, 1.25, 5.0 or 10.0 μmol/kg) or ABT-431 (0.63, 1.25, 2.5 or 5.0 μmol/kg) and were immediately placed in clear plastic observation cages (28 × 17.5 × 13 cm) containing animal bedding and equipped with wire cage lids. Each cage held up to four mice. The animals were observed for behavioral signs of seizure activity that included: buccal movements (rhythmic opening and closing of the mouth), forelimb clonus (repetitive rhythmic clawing motion by one or both of the forepaws that may progress to animals adopting an upright position with arched back and tilted head), clonic seizure, tonic seizure, and death. Bouts of running typically preceded these seizure-like events.

2.4.2. Antagonism of seizures with SCH 23390

Seizure liability of a given dose of A-86929 was determined in the presence of various doses of the dopamine D_1 receptor antagonist, SCH 23390 (0.01, 0.03, 0.10 and 0.30 μmol/kg, s.c.). SCH 23390 or vehicle was injected 5 min prior to administration of a dose (20 μmol/kg, s.c.) of A-86929 that produced seizure activity in 100% of mice as determined in a previous experiment (data not shown).

2.4.3. Effect of subchronic dosing of A-86929 on seizure threshold

The effects of repeated administration of A-86929 on seizure threshold were examined in mice. Animals were administered either vehicle or A-86929 (1.0 μmol/kg, s.c.) once daily for 4 consecutive days. The 1.0 μmol/kg dose of A-86929 was determined from a previous acute dose-response study to be the highest dose that failed to induce any behavioral seizure activity. On day 5, various doses of A-86929 (1.0, 3.0, 7.0, 10, 20 and 30 μmol/kg, s.c.) were administered to mice in each of the two groups. The mice were observed for 2 h for the presence of forelimb clonic seizures.

2.5. Behavioral seizure activity in rats

2.5.1. Acute dose response

For the dose-response determinations, animals were placed into clear plastic observation cages (20 × 10.5 × 8 in) with stainless-steel wire covers. After a 20 min habituation period, young rats (35–37 days old, 90–120 g) were injected with vehicle or varying doses of A-86929 (3.0, 10, 30, 60 and 100 μmol/kg, s.c.) or ABT-431 (3.0, 6.0, 10, 30, 60 and 100 μmol/kg, s.c.) and then returned to the testing cage. Behavior was monitored visually for 3–6 h for signs of excessive excitation (grooming, wet-dog shakes, hyperactivity, pronounced salivation, teeth chattering) and overt seizure activity, typically consisting of rearing and clonic movements of the forelimbs. Each treatment group consisted of 10–14 rats.

Adult rats (3 months old, ~ 350 g) were injected with various doses of A-86929 (3.0, 10.0, 20.0, 60.0 and 100 μmol/kg, s.c.) 60 min after placement into the observation cage. Animals were observed for 5 h for signs of seizure activity. Each treatment group consisted of 8–16 rats.

2.5.2. Antagonism of A-86929-induced seizures

Young rats (35–37 days old, 90–120 g) were allowed a 60-min habituation period in the observation cages and administered either haloperidol (2.76 μmol/kg in 2 ml/kg, s.c.) 20 min prior to A-86929 treatment (100 μmol/kg, s.c.) or SCH 23390 (0.03, 0.09 or 0.31 μmol/kg in 1 ml/kg, s.c.) 5 min prior to A-86929 (100 μmol/kg, s.c.) treatment. Ten rats were used in each treatment group. The animals were observed for 3–6 h for signs of seizure activity.

2.6. Statistics

Analysis of locomotor activity scores was conducted using appropriate analysis of variance (ANOVA) and post-hoc tests. ED_{50} values and 95% confidence limits were determined using the Litchfield and Wilcoxon test (Tallarida and Murray, 1987). Antagonist effects were evaluated by converting the proportions of animals seizing to *Z* scores (Hodges et al., 1975).

3. Results

3.1. Increased locomotor activity in rats

3.1.1. Acute dose response

To establish the presence of a dose-response relation, a one-way ANOVA conducted on total locomotor activity

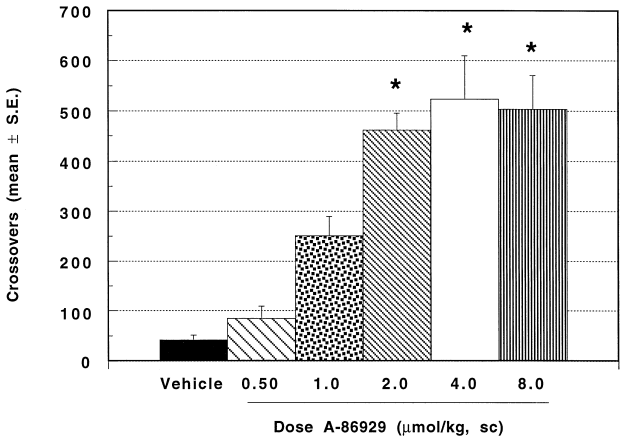


Fig. 2. Locomotor activity, as measured by number of crossovers across a 6 h period, demonstrated by rats after treatment with vehicle or various doses of A-86929. *, significantly different from vehicle; $P < 0.001$.

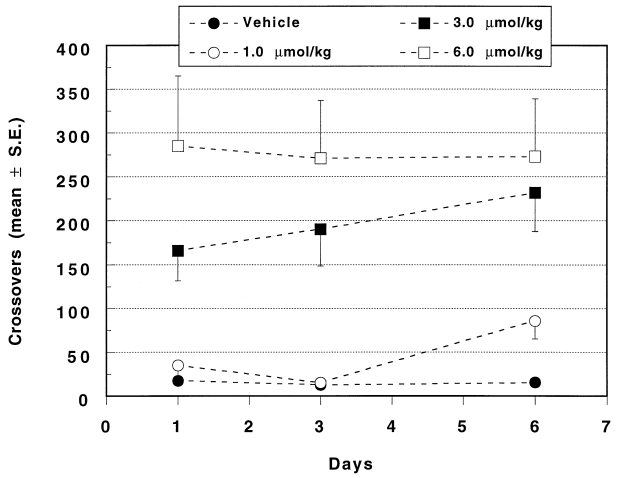


Fig. 3. Locomotor activity on days 1, 3 and 6 in response to daily treatments with vehicle or A-86929 at doses of 1.0, 3.0 and 6.0 μmol/kg, s.c., for 6 days to rats.

scores in response to vehicle or A-86929 indicated significant effects of dose ($F(5,42) = 18.15$; $P < 0.001$) (Fig. 2). Post-hoc analysis (Tukey's HSD) indicated that doses of 2.0, 4.0 and 8.0 μmol/kg, s.c., significantly ($P < 0.001$) increased activity compared to vehicle. Animals treated with 1.0 μmol/kg A-86929 also tended to be hyperactive ($P < 0.06$).

3.1.2. Antagonism of hyperactivity

A one-way ANOVA was conducted on total locomotor activity scores in response to a given dose of A-86929 (1.0 μmol/kg, s.c.) in animals pretreated with varying doses of SCH 23390 (Table 1). This analysis indicated significant effects of dose ($F(4,47) = 15.59$; $P < 0.001$) and post-hoc tests indicated that the two highest doses of SCH 23390 (0.01 and 0.10 μmol/kg, s.c.) significantly reduced locomotor activity ($P < 0.001$) produced by A-86929 compared to vehicle.

Similar analyses were conducted on locomotor activity scores in response to a given dose of A-86929 (1.0 μmol/kg, s.c.) in rats pretreated with various doses of haloperidol (Table 1). There was a significant effect of dose ($F(4,47) = 9.16$; $P < 0.001$), and significant reductions in activity ($P < 0.001$) were produced by the two highest doses of haloperidol (0.1 and 1.0 μmol/kg, s.c.) compared to vehicle.

Table 1
Effects of pretreatment by vehicle or varying doses of SCH 23390 or haloperidol on the locomotor activity elicited by a 1.0 μmol/kg, s.c., dose of A-86929 in rats

Pretreatment	Dose (μmol/kg, s.c.)	Crossovers (mean ± S.E.)
Vehicle		215 ± 28
SCH 23390	0.001	172 ± 26
SCH 23390	0.003	148 ± 28
SCH 23390	0.01	44 ± 8 [a]
SCH 23390	0.10	8.5 ± 1.8 [a]
Haloperidol	0.01	174 ± 44
Haloperidol	0.03	145 ± 39
Haloperidol	0.10	43 ± 5 [a]
Haloperidol	1.00	9 ± 7 [a]

Statistical significance of the difference from vehicle-treated group is indicated as [a] $P < 0.001$.

3.1.3. Effects of subchronic A-86929 treatment on locomotor activity

Analysis of data from the repeated A-86929 treatment study was performed using a two-way ANOVA (dose × days), with repeated measures on the days factor, on total locomotor activity scores on days 1, 3 and 6. A significant effect of dose ($F(4,43) = 14.48$; $P < 0.001$) was obtained, but no significant effect of days ($F(2,8) = 1.32$; $P > 0.25$) or significant dose × day interaction ($F(8,86) = 1.66$; $P > 0.10$) was revealed. These results indicate that although significant hyperactivity was seen with the two higher doses (3.0 and 6.0 μmol/kg, s.c.), the degree of hyperactivity did not change across the testing days (Fig. 3).

3.2. Production of behavioral seizure activity in mice

3.2.1. Acute dose-response study

Varying doses of A-86929 administered subcutaneously produced behavioral seizures in intact mice in a dose-dependent manner (Fig. 4). An ED_{50} value of 7.1 (5.9–9.7) μmol/kg was calculated from these data. Subcutaneous administration of varying doses of ABT-431 produced a similar dose-response curve (Fig. 4), with a corresponding ED_{50} value of 2.7 (1.6–4.5) μmol/kg. Analysis of the doses of A-86929 and ABT-431 that produced behavioral seizure activity in mice indicated no significant difference between the ED_{50} values ($P > 0.05$). The latency to seize was roughly 30–40 min for both compounds.

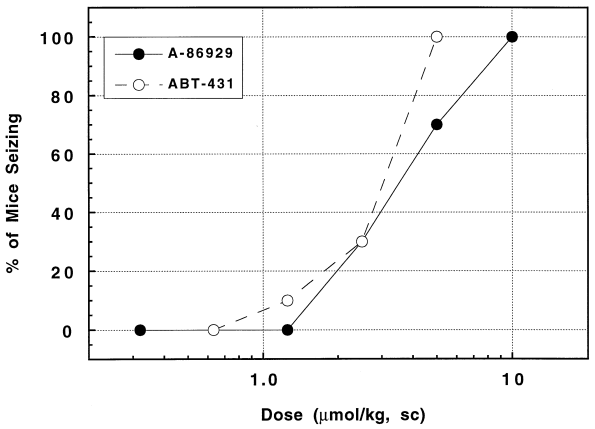


Fig. 4. Dose-response curves of A-86929 and ABT-431 for the production of seizure activity in mice.

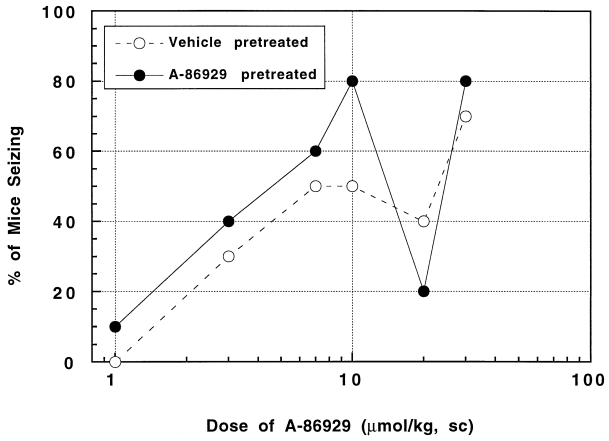


Fig. 5. Effects of 4-day pretreatment with either vehicle or A-86929 (1.0 μmol/kg, s.c.) on dose-response curve of A-86929 to produce seizure activity in mice.

3.2.2. Antagonism of seizures with SCH 23390

The incidence of behavioral seizure activity elicited by A-86929 (20 μmol/kg, s.c., a dose that produced seizures in 100% of the animals in a previous experiment) was reduced in a dose-dependent manner with the dopamine D_1 receptor-selective antagonist SCH 23390 (Table 2). The highest dose of SCH 23390 (0.3 μmol/kg, s.c.) completely protected the mice from seizures induced by A-86929. An ED_{50} value of 0.03 (0.01–0.07) μmol/kg was calculated from these data for the ability of SCH 23390 to antagonize the seizures produced by a 20 μmol/kg dose of A-86929.

3.2.3. Effect of subchronic dosing of A-86929 on threshold of seizure production

Mice that had been pretreated once daily for 4 days with vehicle exhibited behavioral seizure activity in response to various doses A-86929 with an ED_{50} value of 12.8 (5.9–27.8) μmol/kg, s.c. Mice that had been pretreated once daily for 4 days with A-86929 (1.0 μmol/kg, s.c.) also exhibited seizures in response to various doses of A-86929 with an ED_{50} value of 8.4 (1.0–70.9) μmol/kg, s.c. The ED_{50} values generated from the two different groups of animals were not statistically different from each other (Fig. 5). One mouse that had been pretreated for 4 days with A-86929 died during a seizure episode induced after administration of 30 μmol/kg of A-86929 in the subsequent dose-response study. No deaths occurred in mice pretreated with vehicle following administration of various doses of A-86929.

Table 2
Effects of pretreatment by vehicle or varying doses of SCH 23390 on the percentage of mice exhibiting behavioral seizure activity within 3 h following a dose of A-86929 (20 μmol/kg, s.c.)

Dose (μmol/kg, s.c.)	Seizures (% animals)
0.00	100
0.01	88
0.03	50 [a]
0.10	12 [a]
0.30	0 [a]

Statistical significance of the difference from vehicle-treated group is indicated as [a] $P < 0.05$.

3.3. Production of behavioral seizure activity in rats

3.3.1. Acute dose response

A-86929 produced dose-dependent increases in the incidence of behavioral seizure activity in young rats (35–37 days old, 90–120 g) (Fig. 6). The ED_{50} dose in this group of animals was calculated to be 34.2 (19.9–58.8) μmol/kg, s.c., with a mean latency period of 106 min. ABT-431 also produced seizure-like activity in juvenile rats, with an ED_{50} value of 35.6 (20.7–61.4) μmol/kg, s.c., and a mean latency period of 140 min. There was no significant difference between these ED_{50} values.

The incidence of behavioral seizure activity was recorded for various doses of A-86929 administered to adult rats (3 months old, ~ 350 g). An ED_{50} value of 345

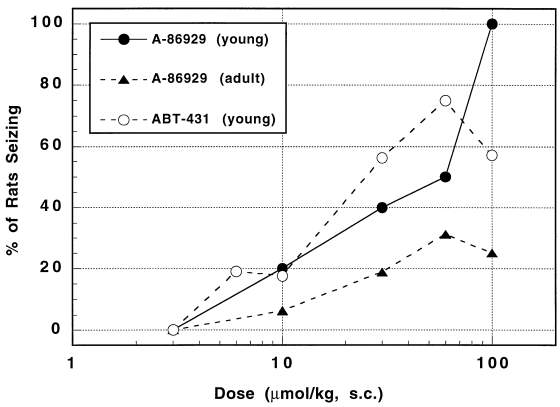


Fig. 6. Dose-response curves of A-86929 and ABT-431 for the production of seizure activity in young (35–37 days old, 90–120 g) rats, and dose-response curve of A-86929 for production of seizure activity in adult (3 months old, ~ 350 g) rats.

Table 3
Effect of selective dopamine D_1 antagonist (SCH 23390) and D_2 antagonist (haloperidol) pretreatment on the incidence of seizure-like activity in rats following injection of A-86929 (100 μmol/kg, s.c.)

Treatment	Dose (μmol/kg, s.c.)	Seizures (% animals)
Vehicle	–	80
SCH 23390	0.03	30 [a]
SCH 23390	0.09	10 [a]
SCH 23390	0.31	10 [a]
Haloperidol	2.7	40a

Each of the treatment groups consisted of ten animals. Statistical significance of the difference from the vehicle-treated group is indicated as [a] $P < 0.05$.

(126–940) μmol/kg, s.c., was calculated for this group (Fig. 6). This difference in ED_{50} values for A-86929-induced seizure incidence between the young and adult rat groups was statistically significant ($P < 0.05$).

3.3.2. Antagonism of A-86929-induced seizure activity in rats

Analysis of the antagonist data showed that all pretreatment doses (0.03, 0.09 and 0.31 μmol/kg, s.c.) of SCH 23390 significantly reduced the incidence of seizure activity produced by a given dose of A-86929 (100 μmol/kg, s.c.) compared to vehicle pretreatment (Table 3). Haloperidol also significantly reduced the number of rats exhibiting seizure-like activity but at a higher dose ($P < 0.05$ for all comparisons). Latencies to seizure onset were increased by both antagonists (data not shown).

4. Discussion

A-86929 produced a dose-dependent increase in horizontal locomotor activity in neurologically intact rats. The A-86929-induced locomotor effects are consistent with those reported for other dopamine D_1 receptor-selective full agonists such as SKF 89626 (4(3′,4′-dihydroxyphenyl)-4,5,6,7-tetrahydrothieno(2,3-*c*)-pyridine) (Andersen et al., 1987) and the benzazepine SKF 82958 (Meyer and Shults, 1993). In contrast, the dopamine D_1 receptor partial agonist SKF 38393 was reported to have very weak or no ability to increase horizontal locomotor activity following either systemic or direct intracerebral administration (Andersen et al., 1987; Meyer and Shults, 1993). Rather, the partial agonist has been reported to produce grooming, sniffing, vacuous chewing and other stereotyped behaviors in rats (Murray and Waddington, 1989). Specific monitoring for these behaviors was not conducted in the present study.

The A-86929-induced hyperactivity could be attenuated in a dose-dependent manner by the selective dopamine D_1 receptor antagonist SCH 23390 at doses ≥ 0.01 μmol/kg, s.c., indicating the involvement of dopamine D_1 receptor stimulation in this behavior. However, the preferential dopamine D_2 receptor antagonist, haloperidol, also attenuated the A-86929-induced hyperactivity to roughly the same extent as that produced by SCH 23390, although higher doses were required (≥ 0.10 μmol/kg, s.c.). The ability of a dopamine D_2 receptor antagonist to greatly attenuate the D_1 agonist-induced hyperactivity may reflect functional interactions between dopamine D_1 and D_2 receptors that have been reported in vitro and in vivo (Jackson and Westlind-Danielsson, 1994). For example, episodic increases in locomotor activity produced by administration of SKF 38393 to intact rats were reported to be antagonized by SCH 23390 as well as by higher doses of the dopamine D_2 receptor antagonist metoclopramide. Likewise, the increased locomotor activity and stereotyped behaviors produced by dopamine D_2 receptor-selective agonists could be effectively reversed by D_1-selective antagonists in neurologically intact rats (Molloy and Waddington, 1985; Pugh et al., 1985). In rats with unilateral dopamine depletions, the actions of SKF 38393 were selectively blocked by dopamine D_1, but not D_2, receptor antagonists (Arnt, 1985), suggesting a change in receptor subtype interactions after chemical lesioning. Others have reported synergistic effects of dopamine D_1 plus D_2 receptor agonist treatments in 6-hydroxydopamine-lesioned rats (Robertson and Robertson, 1986).

In the current study, constant levels of hyperactivity were maintained following daily administration of three different doses of A-86929 to intact rats for 6 days. This maintenance of behavioral responsivity is consistent with that observed following once or three-times daily administration of varying doses of A-86929 for 10 consecutive days to 6-hydroxydopamine-lesioned rats (Michaelides et al., 1995) and to MPTP-lesioned marmosets (Shiosaki et al., 1996). Maintaining behavioral responsivity with repeated administration by a dopamine receptor agonist will be important in establishing whether a particular compound will have potential as chronic therapy. Rapid behavioral tolerance was seen with repeated administration of the long-acting isochroman dopamine D_1 receptor agonist, A-77636 ((1*R*,3*S*) 3-(1′-adamantyl)-1-aminomethyl-3,4-dihydro-5,6-dihydroxy-1*H*-2-benzopyran hydrochloride) (Asin and Wirtshafter, 1993) or with constant infusion of SKF 38393 (Winkler and Weiss, 1989) to 6-hydroxydopamine-lesioned rats. Prolonged stimulation of the dopamine D_1 receptor with a long-acting compound or by continuous infusion may contribute to the behavioral tolerance observed. However, the relation between dosing regimen and behavioral responsivity following dopamine agonist treatment is very complex (Post, 1980), and both sensitization and tolerance have both been reported after repeated administration of selective dopamine D_1 and D_2 receptor agonists to rats (c.f.: Braun and Chase, 1988; Mattingly et al., 1993; Neisewander et al., 1991; White et al., 1990).

Acute administration of A-86929 and ABT-431 to mice

produced behavioral seizure activity in a dose-dependent manner, with ED_{50} values of 7.1 (5.9–9.7) and 2.7 (1.6–4.5) μmol/kg, s.c., respectively. The lack of a significant difference in the behavioral potencies of the two compounds supports a rapid conversion in vivo of both acetyl groups in ABT-431 to the parent catechol A-86929 following subcutaneous administration. This is consistent with a previous report in which no difference was observed in the potencies of A-86929 and ABT-431 for inducing contralateral rotation following subcutaneous administration to 6-hydroxydopamine-lesioned rats (Shiosaki et al., 1996). The behavioral seizures produced by A-86929 were qualitatively similar to those reported in mice following treatment with the structurally dissimilar dopamine D_1 receptor agonists, A-68930 and SKF 82958 (Britton et al., 1991). The A-86929-induced seizures were blocked in a dose-dependent manner by SCH 23390, indicating the involvement of dopamine D_1 receptor stimulation in this behavior. Pretreating animals with a subthreshold dose (the highest no-seizure dose) of A-86929 did not affect the seizure threshold when rats were subsequently challenged with higher doses of the compound. This result is consistent with our failure to observe either behavioral sensitization or tolerance in the locomotor activity studies with repeated A-86929 treatments.

Both A-86929 and ABT-431 produced behavioral seizure activity with similar potencies (ED_{50} = 34.2 and 35.6 μmol/kg, s.c., respectively) in young rats weighing approximately 100 g. In comparison to mice, rats were less sensitive to the seizure-inducing properties of A-86929 and ABT-431, and their latency to seize was longer. Interestingly, the incidence of seizures induced by A-86929 was reduced by either SCH 23390 or haloperidol pretreatment. In other studies, haloperidol generally has been found to lower seizure thresholds in response to a variety of electrical and pharmacological stimuli (see review in Starr, 1996). Of particular relevance to the present study was the report that haloperidol enhanced the ability of the dopamine D_1 receptor agonist, CY 208–243, to promote pilocarpine-induced seizures in mice (Burke et al., 1990). However, the seizures observed in the present study were in response to treatment with a D_1 agonist alone, and, in this case, haloperidol effectively attenuated the incidence of behavioral seizures. The ability of haloperidol to block the seizures induced by A-86929 may be a logical continuum of its ability to block the locomotor stimulant actions of this agonist, and may further speak to the issue of dopamine D_1/D_2 receptor interactions in the neurologically intact animal.

The effects of A-86929 on the production of behavioral seizure activity were also studied in adult rats weighing approximately 350 g. In this group of animals, the dose-response curve produced by A-86929 was more shallow than that produced in younger rats. An ED_{50} value of 345 μmol/kg, s.c., was calculated from these data and this value is ten-fold higher than that generated from young rats (ED_{50} = 34.2 μmol/kg, s.c.). The reasons underlying this difference in sensitivity are not understood. Age-dependent differences in D_1 receptor levels between the brain of a young rat (35–37 days) relative to that of an adult rat (3 months) are unlikely to contribute to the difference in seizure sensitivity (Rao et al., 1991). The adults rats used in these studies were approximately three-times heavier than the young rats, and differences in body fat content in the adults may contribute to differences in the distribution and pharmacokinetics of A-86929 relative to younger rats and may, in part, account for the difference in seizure sensitivities.

A comparison of the ED_{50} values for A-86929 that produced behavioral seizure activity in rats (ED_{50} = 34.2 (juvenile) and 345 (adult) μmol/kg, s.c.) with the previously established ED_{50} value (ED_{50} = 0.24 μmol/kg, s.c.) for A-86929 that produced contralateral rotation in the 6-hydroxydopamine-lesioned rat model of Parkinson's disease (Shiosaki et al., 1996) indicates a significant dose window separating the production of these behavioral effects produced by this compound. Characterization of the behavioral effects associated with increasing the doses of A-86929 and, by extension, ABT-431, in preclinical models is an important consideration in advancing compounds for future clinical study.

References

Al-Tajir, G., C. Chander, B. Starr and M. Starr, 1990a, Opposite effects of stimulation of D1 and D2 dopamine receptors on the expression of motor seizures in mouse and rat, Neuropharmacology 29, 657.

Al-Tajir, G., M. Starr and B. Starr, 1990b, Proconvulsant effect of SKF 38393 mediated by nigral D1 receptors, Eur. J. Pharmacol 182, 245.

Andersen, P.H., E.B. Nielsen, J. Scheel-Kruger, J.A. Jansen and R. Hohlweg, 1987, Thienopyridine derivatives identified as the first selective, full efficacy, dopamine D1 receptor agonists, Eur. J. Pharmacol. 137, 291.

Arnt, J., 1985, Behavioural stimulation is induced by separate dopamine D-1 and D-2 receptor sites in reserpine-pretreated but not in normal rats, Eur. J. Pharmacol. 113, 79.

Asin, K. and D. Wirtshafter, 1993, Effects of repeated dopamine D1 receptor stimulation on rotation and c-fos expression, Eur. J. Pharmacol. 235, 167.

Barone, P., S.A. Parashos, V. Palma, C. Marin, G. Campanella and T. Chase, 1990, Dopamine D1 receptor modulation of pilocarpine-induced convulsions, Neuroscience 34, 209.

Braun, A. and T. Chase, 1988, Behavioral effects of chronic exposure to selective D-1 and D-2 dopamine receptor agonists, Eur. J. Pharmacol. 147, 441.

Britton, D.R., K.E. Asin, L. Yahiro, P. Curzon, L. Bednarz and J. Williams, 1991, D1 dopamine receptor agonists induce forelimb clonus [abstract], Soc. Neurosci. 17, 638.

Burke, K., C.J. Chandler, B.S. Starr and M.S. Starr, 1990, Seizure promotion and protection by D-1 and D-2 dopaminergic drugs in the mouse, Pharmacol. Biochem. Behav. 36, 729.

Eilam, D., H. Talangbayan, G. Canaran and H. Szechtman, 1992, Dopaminergic control of locomotion, mouthing, snout contact, and grooming: opposing roles of D1 and D2 receptors, Psychopharmacology 106, 447.

Hodges, J.L., D. Krech and R. Crutchfield, 1975, Statlab: An Empirical Introduction to Statistics (McGraw-Hill, New York, NY) p. 185.

Jackson, D. and A. Westlind-Danielsson, 1994, Dopamine receptors: Molecular biology, biochemistry and behavioural aspects, Pharmacol. Ther. 64, 291.

Jenner, P., S. Rose, M. Nomoto and C. Marsden, 1986, MPTP-induced parkinsonism in the common marmoset: Behavioral and biochemical effects, Adv. Neurol. 45, 183.

Mattingly, B., J. Rowlett and G. Lovell, 1993, Effects of daily SKF 38393, quinpirole, and SCH 23390 treatments on locomotor activity and subsequent sensitivity to apomorphine, Psychopharmacology 110, 320.

Meyer, M. and J. Shults, 1993, Dopamine D1 receptor family agonists, SK&F38393, SK&F77434, and SK&F82958, differentially affect locomotor activities in rats, Pharmacol. Biochem. Behav. 46, 269.

Michaelides, M.R., Y. Hong, S.J. DiDomenico, K.E. Asin, D.R. Britton, C.W. Lin, M. Williams and K. Shiosaki, 1995, (5*aR*,11*bS*)-4,5,5*a*,6,7,11*b*-Hexahydro-2-propyl-3-thia-5-azacyclopent-1-ena[*c*]-phenanthrene-9,10-diol (A-86929): a potent and selective dopamine D1 agonist that maintains behavioral efficacy following repeated administration, and characterization of its diacetyl prodrug (ABT-431), J. Med. Chem. 38, 3445.

Molloy, A. and J. Waddington, 1985, The enantiomers of SK&F 83566, a new selective D-1 dopamine antagonist, stereospecifically block stereotyped behaviour induced by apomorphine and by the selective D-2 agonist RU 24213, Eur. J. Pharmacol. 116, 183.

Molloy, A., K. O'Boyle, M. Pugh and J. Waddington, 1986, Locomotor behaviors in response to new selective D-1 and D-2 dopamine receptor agonists, and the influence of selective antagonists, Pharmacol. Biochem. Behav. 25, 249.

Murray, A.M. and J.L. Waddington, 1989, The induction of grooming and vacuous chewing by a series of selective D-1 dopamine receptor agonists: two directions of D-1:D-2 interaction, Eur. J. Pharmacol. 160, 377.

Neisewander, J., I. Lucki and P. McGonigle, 1991, Behavioral and neurochemical effects of chronic administration of reserpine and SKF-38393 in rats, J. Pharmacol. Exp. Ther. 257, 850.

Ogren, S. and B. Pakh, 1993, Effects of dopamine D1 and D2 receptor agonists and antagonists on seizures induced by chemoconvulsants in mice, Pharmacol. Toxicol. 72, 213.

Post, R., 1980, Intermittent versus continuous stimulation: Effect of time interval on the development of sensitization or tolerance., Life Sci. 26, 1275.

Pugh, M., M. O'Boyles, A. Molloy and J. Waddington, 1985, Effects of the putative D-1 antagonist SCH 23390 on stereotyped behavior induced by the D-2 agonist Ru 24213, Psychopharmacology 87, 308.

Rao, P.A., P.B. Molinoff and J.N. Joyce, 1991, Ontogeny of dopamine D1 and D2 receptor subtypes in rat basal ganglia: a quantitative autoradiographic study, Brain Res. Dev. Brain Res. 60, 161.

Robertson, G. and H. Robertson, 1986, Synergistic effects of D1 and D2 dopamine agonists on turning behaviour in rats, Brain Res. 384, 387.

Shiosaki, K., P. Jenner, K. Asin, D. Britton, C. Lin, M. Michaelides, L. Smith, B. Bianchi, S. DiDomenico, L. Hodges, Y. Hong, L. Mahan, J. Mikusa, T. Miller, A. Nikkel, M. Stashko, D. Witte and M. Williams, 1996, ABT-431: The diacetyl prodrug of A-86929, a potent and selective dopamine D1 receptor agonist: In Vitro characterization and effects in animal models of parkinson's disease, J. Pharmacol. Exp. Ther. 276, 150.

Starr, M.S., 1996, The role of dopamine in epilepsy, Synapse 22, 159.

Tallarida, R.J. and R.B. Murray, 1987, Manual of pharmacological calculations with computer programs (Springer, New York, NY) p. 153.

Turski, W., E. Cavalheiro, C. Ikonomidou, Z. Bortolotto, T. Klockgether and L. Turski, 1990, Dopamine control of seizure propagation: intranigral dopamine-D1 agonist SKF-38393 enhances susceptibility to seizures, Synapse 5, 113.

White, F., H. Xiu-Ti and R. Brooderson, 1990, Repeated stimulation of dopamine D1 receptors enhances the effects of dopamine receptor agonists, Eur. J. Pharmacol. 191, 497.

Winkler, J. and B. Weiss, 1989, Effect of continuous exposure to selective D1 and D2 dopaminergic agonists on rotational behavior in supersensitive mice, J. Pharmacol. Exp. Ther. 249, 507.