Pergamon

Pharmacology Biochemistry and Behavior, Vol. 47, No. 3, pp. 753–756, 1994

0091-3057/94 $6.00 + .00

BRIEF COMMUNICATION

Behavioral Profile of Raclopride in Agonistic Encounters Between Male Mice

M. A. AGUILAR, J. MIÑARRO, N. PÉREZ-IRANZO AND V. M. SIMÓN[1]

Area de Psicobiología, Facultad de Psicología,
Universitat de València. Apartado 22109, 46071 Valencia, Spain

Received 26 April 1993

AGUILAR, M. A., J. MIÑARRO, N. PÉREZ-IRANZO AND V. M. SIMÓN. *Behavioral profile of raclopride in agonistic encounters between male mice.* PHARMACOL BIOCHEM BEHAV 47(3) 753–756, 1994.—Raclopride is a substituted benzamide with high selectivity as an antagonist of central dopaminergic D_2 receptors and potential antipsychotic effects. In comparison with a classic DA receptor blocking agent like haloperidol, raclopride displays an atypical profile in preclinical tests for extrapyramidal side effects. Antiaggressive properties of raclopride on agonistic behavior have not yet been fully explored. In this work the effects of raclopride (0.1, 0.3, or 0.6 mg/kg) on aggressive and motor behaviors in male mice were studied. Aggression tests were performed 30 min after injections. Encounters were videotaped and behavior was evaluated, measuring the time spent in 11 broad categories of behavior. The results show a clear antiaggressive effect of raclopride, with very little motor impairment and some increase in exploratory behavior. This behavioral profile is very similar to the one observed with other atypical neuroleptics and differs somewhat from that found in the classic compounds.

Raclopride Aggression Agonistic behavior Mice

RACLOPRIDE is a substituted benzamide that acts as a dopamine antagonist, showing a high selectivity for central dopaminergic D_2 receptors (4) and possessing potential antipsychotic effects. In comparison with a classic DA receptor blocking agent, like haloperidol, raclopride displays an atypical profile in preclinical tests for extrapyramidal side effects (7,10,16). Comparatively high doses of raclopride are needed to induce catalepsy, and its potency to affect locomotor activity, treadmill locomotion, and a conditioned avoidance response is half that of haloperidol (7). Similarly, Ögren et al. (16) found that raclopride was less potent than haloperidol in blocking the apomorphine-induced stereotypies (chewing, biting, licking); however, raclopride and haloperidol were almost equally potent in inhibiting the increase in locomotion caused by apomorphine.

The antiaggressive properties of neuroleptic drugs were known long ago (6,8), but the effects of the recently developed compound raclopride on agonistic behavior have not been fully explored. The chemical structure of raclopride is similar to that of sulpiride, but raclopride has the advantage that it readily penetrates the blood–brain barrier, reaching high concentrations in the brain shortly after injection (9). In view of the similarity of its chemical structure to sulpiride, it was hypothesized that raclopride would show a similar behavioral profile on animal aggression models. In the isolation-induced aggression paradigm, sulpiride reduces aggressive behavior at doses producing little motor impairment (17). Other atypical neuroleptics, such as clozapine, have also shown a similar pattern of activity (5).

In the present work, an attempt was made to explore the acute effects of raclopride on aggressive behavior, using a range of doses. This range is similar to that used by researchers in numerous behavioral experiments with rats: self-administration of heroin (13); study of various motor behaviors

[1] To whom requests for reprints should be addressed.

(7,19); brain stimulation and food reward (14). Aggressive behavior was studied in isolated male mice confronting anosmic conspecifics.

METHOD

Subjects

Ninety albino male mice of the OF1 strain acquired in IFFA CREDO (Barcelona) were used in this study. Subjects arrived in the laboratory at 42 days of age and were housed under standard conditions: constant temperature (21°C), a reversed light schedule (white lights on: 0100–1300 h), and food and water available ad lib, except during behavioral tests. Half of the animals were individually housed in transparent plastic cages (24 × 13.5 × 13 cm) and were used as experimental and control animals. The remainder were housed in groups of six to be used as "standard opponents" and were made temporarily anosmic by intranasal lavage with 4% zinc sulphate solution on both 1 and 3 days before testing (18). This kind of opponent was used because it elicits attack but never initiates such behavior (2). All animals underwent an adaptation period of 30 days before experimental treatments were applied.

Experimental Design

Individually housed animals were randomly allocated to one control group (N = 12) receiving physiological saline and three experimental groups (N = 11 each) receiving raclopride injections. All animals (individually housed as well as standard opponents) were used only once.

Drug Treatment

Animals were injected IP with three doses (0.1, 0.3, or 0.6 mg/kg) of raclopride tartrate (batch F7, Astra Lab., Södertälje, Sweden) in a volume of 0.01 ml/g. A control group was injected with physiological saline. Raclopride was also dissolved in the same solution. Aggression tests were performed 30 min after injections.

Social Encounters

After injections an experimental animal and a standard opponent (marked with fur dye for identification) confronted each other in a neutral cage for 10 min. The animals were allowed 1 min of adaptation to the neutral cage, whilst separated by means of a plastic barrier before the encounter. This neutral cage consisted of an all-glass area, measuring 60 × 33 × 30 cm. Encounters were videotaped using a Panasonic VHS camera. All tests were carried out under white illumination between the second and fifth hour of the dark phase of the light/dark cycle. After each encounter the neutral cage was washed out and the sawdust bedding replaced.

Behavioral Analysis

The tapes were analyzed using a microprocessor (Commodore 64 computer) and a custom-developed program (3) that facilitated estimation of times allocated to 11 broad behavioral categories. Each category included a collection of different behavioral postures and elements. The names of categories and their constituent elements are as follows:

1. body care (abbreviated groom, self-groom, wash, shake, scratch);
2. digging (dig, kick dig, push dig);
3. nonsocial exploration (explore, rear, supported rear, scan);
4. explore from a distance (approach, attend, circle, head orient, stretched attention);
5. social investigation (crawl over, crawl under, follow, groom, head groom, investigate, nose sniff, sniff, push past, walk around);
6. threat (aggressive groom, sideways offensive, upright offensive, tail rattle);
7. attack (charge, lunge, attack, chase);
8. avoidance/flee (evade, flinch, retreat, ricochet, wheel, startle, jump, leave, wall clutch);
9. defensive/submissive (upright defensive, upright submissive, sideways defensive);
10. sexual (attempted mount, mount);
11. immobility (squat, cringe).

A detailed description of all elements can be found in Brain et al. (3) and Martinez et al. (11). The analysis of the videotape involved assessment only of the behavior of the experimental and control animals. This analysis was performed by a trained experimenter who was blind as to the experimental group to which each animal belonged.

Statistical Analysis

The medians for times allocated to each broad behavioral category were determined. Nonparametric Kruskal–Wallis tests were used to assess the variance of the behavioral measures over different treatment groups. Subsequently, appropriate paired comparisons were carried out using Mann–Whitney U-tests, to contrast the behavior in different treatment groups.

RESULTS

Table 1 illustrates medians (with ranges) of accumulated times allocated to the broad categories of behavior described above. Kruskal–Wallis analysis showed that there was significant variance in the category of attack ($p < 0.004$). In comparison with the saline group, all doses of raclopride (0.1, 0.3, and 0.6 mg/kg) significantly reduced time allocated to attack behavior ($p < 0.02$).

DISCUSSION

In this study, raclopride produced a very clear dose-dependent antiagressive effect, reducing attack behavior to very low levels, even almost abolishing it with the highest dose of 0.6 mg/kg. It must be emphasized that this effect reached significance in the three explored doses. Although no other behavioral category in this study showed significant changes, it might be of interest to point out that there was an apparent dose-dependent increase in nonsocial exploration and no change in immobility was observed. On the other hand, threat behavior, the other behavioral category related to aggression, was reduced only by the highest dose, although not significantly.

Animals treated with the dose range, which significantly reduced attack behavior, showed no apparent motor impairment. With reference to this, the category of immobility showed only a slight increase with the highest dose, but without reaching significance (a median of 3 s was measured, out of a total duration of 600 s). Other behaviors with an evident motor component, such as the three exploratory categories—nonsocial exploration, explore from a distance, and social investigation—did not exhibit significant decreases. In fact, the category of nonsocial exploration exhibited a nonsignificant increase.

TABLE 1

MEDIAN VALUES (WITH RANGES) FOR TIMES (IN s) ALLOCATED TO BROAD BEHAVIORAL CATEGORIES IN ANIMALS RECEIVING ONE SINGLE INJECTION OF RACLOPRIDE

		Doses of Raclopride		
Behavioral Category	Saline (n = 12)	0.1 mg/kg (n = 11)	0.3 mg/kg (n = 11)	0.6 mg/kg (n = 11)
Body care	14 (0–38)	24 (3–46)	19 (8–44)	29 (9–55)
Digging	7 (0–25)	6 (0–26)	8 (1–30)	1 (0–35)
Nonsocial exploration	282 (220–418)	319 (163–396)	325 (122–393)	341 (248–401)
Explore from a distance	62 (10–119)	38 (8–156)	64 (28–132)	60 (18–106)
Social investigation	48 (5–135)	87 (26–218)	98 (16–150)	93 (25–175)
Threat	61 (1–146)	63 (0–147)	75 (2–134)	27 (0–161)
Attack*	69 (0–204)	31† (0–96)	18† (0–93)	0† (0–150)
Avoidance/flee	0 (0–3)	0 (0–1)	0 (0–5)	0 (0–0)
Defensive/submissive	0 (0–0)	0 (0–0)	0 (0–0)	0 (0–0)
Sexual	0 (0–0)	0 (0–0)	0 (0–0)	0 (0–0)
Immobility	0 (0–9)	0 (0–336)	0 (0–210)	3 (0–63)

*Kruskal-Wallis test showed significant variance, $p < 0.004$.
†Differs from controls on two-tailed Mann-Whitney U-test, $p < 0.02$.

A robust inhibition of isolation-induced aggression has been a common finding in experiments involving dopaminergic antagonists and aggressive encounters in male rodents. In fact, all dopaminergic antagonists seem to share strong antiaggressive properties but differ by the amount of motor impairment produced. In experiments carried out using the same techniques, classical neuroleptics like haloperidol (12) or spiperone (1) do produce a considerable amount of motor impairment, whereas atypical ones like sulpiride (17) or clozapine (5) show limited effects on motor behaviors. Raclopride undoubtedly belongs to the group of dopamine antagonists with less effects on motor behavior. The evidence collected to date suggests that the antiaggressive power of dopaminergic antagonists is related to their attenuating actions on reinforcement (14) being quite independent of their motor effects, and that both phenomena could have different underlying neurological mechanisms (15).

ACKNOWLEDGEMENT

We thank ASTRA ARCHUS A.B. for providing the drug used in this work.

REFERENCES

1. Arregi, A.; Azpiroz, A.; Simón, V. M.; Brain, P. F. Effects of two dopaminergic selective antagonists on ethologically assessed conflicts in male mice. Gen. Pharmacol. 24(2):353–356; 1993.
2. Brain, P. F.; Benton, D.; Childs, G.; Parmigiani, S. The effect of the opponent in tests of murine aggression. Behav. Processes 6:319–328; 1981.
3. Brain, P. F.; Mc Allister, K. H.; Walmsley, S. V. Drug effects on social behaviour. Methods in ethopharmacology. In: Boulton, A. A.; Baker, G. B.; Greenshaw, A. J., eds. Neuromethods. Clifton, NJ: The Humana Press Inc.; 1989:687–739.
4. de Paulis, T.; Kumar, Y.; Johansson, L.; Rämsby, S.; Hall, H.; Sällemark, M.; Angeby-Möller, K.; Ogren, S-O. Potential neuroleptic agents. 4. Chemistry, behavioural pharmacology and blocking of 3H-spiroperone binding of 3,5-disubstituted N-[(1-ethyl-2-pyrrolidinyl)methyl]-6-methoxysalicylamide. J. Med. Chem. 29: 61–69; 1986.
5. Garmendia, L.; Sánchez, J. R.; Azpiroz, A.; Brain, P. F.; Simón, V. M. Clozapine: Strong antiaggressive effects with minimal motor impairment. Physiol. Behav. 51:51–54; 1991.
6. Gianutsos, G.; Lal, H. Narcotic analgesics and aggression. Mod. Probl. Pharmacopsychiatry 13:114–138; 1978.
7. Hillegaart, V.; Ahlenius, S. Effects of raclopride on exploratory locomotor activity, treadmill locomotion, conditioned avoidance behaviour and catalepsy in rats: Behavioural profile comparisons between raclopride, haloperidol and preclamol. Pharmacol. Toxicol. 60:350–354; 1987.
8. Janssen, P. A. J.; Jageneau, A. H.; Niemegeers, C. J. E. Effects of various drugs on isolation-induced fighting behavior of male mice. J. Pharmacol. Exp. Ther. 129:471–475; 1960.

9. Köhler, C.; Hall, H.; Ögren, S-O.; Gawell, L. Specific in vitro and in vivo binding of 3H-raclopride: A potent substituted benzamide drug with high affinity for dopamine D-2 receptors in the rat brain. Biochem. Pharmacol. 34:2251–2259; 1985.
10. Ljungberg, T. Attenuation of water intake and operant responding by dopamine D2 antagonists: Raclopride provides important cues for understanding the functional mechanism of action. Pharmacol. Toxicol. 65:9–12; 1989.
11. Martinez, M.; Castaño, D.; Simón, V. M.; Brain P. F. An ethopharmacological assessment of the influences of cyproterone acetate on social interactions in male mice. IRCS Med. Sci. 14:44–45; 1986.
12. Miñarro, J.; Castaño, D.; Brain, P. F.; Simón, V. M. Haloperidol does not antagonize the effects of stress on aggressive behaviour in mice. Physiol. Behav. 47:281–285; 1990.
13. Nakajima S.; Wise, R. A. Heroin self-administration in the rat suppressed by SCH 23390. Soc. Neurosci. Abstr. 429:9; 1987.
14. Nakajima, S.; Baker, J. D. Effects of D2 dopamine receptor blockade with raclopride on intracranial self-stimulation and food reinforced operant behavior. Psychopharmacology (Berlin) 98:330–333; 1989.
15. Navarro, J. F.; Miñarro, J.; Simón, V. M. Antiaggressive and motor effects of haloperidol show different temporal patterns in the development of tolerance. Physiol. Behav. 53:1055–1059; 1993.
16. Ögren, S-O.; Hall, H.; Köhler, C.; Magnusson, O.; Sjöstrand, S-E. The selective dopamine D2 receptor antagonist raclopride discriminates between dopamine-mediated motor functions. Psychopharmacology (Berlin) 90:287–294; 1986.
17. Redolat, R.; Brain, P. F.; Simón, V. M. Sulpiride has an antiaggressive effect in mice without markedly depressing motor activity. Neuropharmacology 30:41–46; 1991.
18. Smoothy, R.; Brain, P. F.; Berry, M. S.; Haug, M. Alcohol and social behaviour in group-housed female mice. Physiol. Behav. 37:689–694; 1986.
19. Wadenberg, M. L.; Ahlenius, S. Antipsychotic-like profile of combined treatment with raclopride and 8-OH-DPAT in the rat: Enhancement of antipsychotic-like effects without catalepsy. J. Neural Transm. 83:43–53; 1991.