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5-HT_{1A} Agonist Effects on Punished Responding of Squirrel Monkeys

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GLEESON, S. AND J. E. BARRETT. *5-HT_{1A} agonist effects on punished responding of squirrel monkeys.* PHARMACOL BIOCHEM BEHAV **37**(2) 335–337, 1990.—Buspirone and other drugs that act as 5-HT_{1A} agonists appear to be clinically effective anxiolytics in humans, yet their anticonflict effects, though robust in pigeons, are equivocal in rodents. In the present study we examined the effects of the benzodiazepine midazolam and a series of 5-HT_{1A} agonists on punished responding of squirrel monkeys. Lever presses were reinforced according to a fixed-interval 3-min schedule; in addition, each thirtieth lever press was punished. Midazolam produced large increases in response rates, whereas none of the 5-HT_{1A} compounds produced any increases in responding. Most of these drugs decreased response rates at the higher doses examined. Although the reasons for the discrepancy between species in the anticonflict effects of serotonergic anxiolytics cannot be specified, the different anatomical distribution of 5-HT_{1A} binding sites across species may suggest a different functional role for this receptor.

5-HT_{1A} agonists Punished responding Anxiolytics Buspirone Midazolam 8-OH-DPAT Ipsapirone
BMY 7378 RU 24969 SM 3997 WY 47,846 Flesinoxan Squirrel monkeys

A number of drugs that show anxiolytic effects in humans (e.g., benzodiazepines, barbiturates) also increase food-maintained responding suppressed by punishment (10) and water-tube licking suppressed by electric shock (22) in nonhumans. These conflict procedures constitute the most common laboratory methods for assessing the antianxiety potential of novel compounds (1). In this regard, substantial experimental attention has focused on drugs that act as agonists at the 5-HT_{1A} receptor (7,20). These drugs are exemplified by buspirone, a clinically effective antianxiety agent (13,21), without the sedative/hypnotic/anticonvulsant effects common to the benzodiazepines.

In contrast to the therapeutic potential of buspirone and related compounds, studies of these drugs in conflict procedures have yielded mixed results (18). For example, of the studies reporting an anticonflict effect, some have obtained smaller increases in responding with 5-HT_{1A} agonists than with benzodiazepines. In other cases, these drugs either have no effect or actually decrease punished responding (3, 6–8). Mixed effects also have been reported in the few studies of buspirone in monkeys. Weissman *et al.* (24) and Geller and Hartmann (9) reported increases in punished responding, whereas Wettstein (25) found that buspirone did not increase punished responding after either acute or chronic administration. However, the clinical potential of 5-HT_{1A} agonists is reflected in their anticonflict effects in pigeons. In this species, buspirone, gepirone, and 8-OH-DPAT increase punished responding to levels similar to that observed with the benzodiazepine chlordiazepoxide (4,17). Large increases in punished responding also have been reported for ipsapirone, BMY 7378, RU 24969 (11), and flesinoxan (2), all of which exhibit affinity for the 5-HT_{1A} receptor site (5, 12, 26).

Reasons for the discrepancy between pigeons and other species in the anticonflict effects of 5-HT_{1A} agonists are not known. Studies using rats have varied greatly in methods, including dose range, route of administration, time course, and details of the behavioral procedures [although systematic manipulation of these variables does not seem to alter the negative effects of buspirone (16)]. Studies in monkeys have been limited to buspirone, which affects catecholamine as well as serotonin neurotransmission. In the present experiment we examined the anticonflict effects in squirrel monkeys of several 5-HT_{1A} agonists that have been shown to be effective in pigeons, as well as two others, WY 47,846 and SM 3997, that have shown some anticonflict activity in rats (15,19).

METHOD

Subjects and Apparatus

Six adult male squirrel monkeys (*Saimiri sciureus*) served as subjects. They were individually housed in a colony room with constant temperature and humidity and a 14-hr light cycle. The monkeys were maintained at approximately 85% of free-feeding weight (800–1050 g) by daily rations of New World Monkey Diet and fruit. Water was continuously available in the home cages. All monkeys had been used in previous behavioral experiments and had received drugs.

Experimental sessions were conducted with the monkey seated in a transparent Plexiglas primate chair (14) placed in a sound- and light-attenuating chamber. A stainless steel response lever (BRS/LVE) requiring a force of approximately 0.2 N to operate was located on the front panel of the chair. A food magazine located behind the front panel delivered 300-mg banana-flavored pellets into a receptacle recessed behind the panel. Red stimulus lamps

were arranged behind the panel at eye level and a relay mounted behind the lever produced a feedback click when the lever was operated. The monkey's tail was held in place by a small stock mounted below the seat. Electrode paste was applied to the shaved distal section of tail on which two brass electrode plates rested. Electrical stimulation from a 650-V AC 60-Hz transformer with output wired through series resistance was delivered through the plates. Intensities were adjusted individually for each animal (1 to 4 mA) and were monitored by an AC meter (Simpson model No. 1257). Shock duration was 200 msec.

Procedure

Responding was maintained by a fixed-interval 3-min (FI 3-min) schedule of pellet delivery, with a 1-min limited hold. That is, the first response after 3 min elapsed produced a food pellet, but the interval was terminated without food if the response did not occur within 1 min after the pellet was available. In addition, every thirtieth response during the FI produced shock. The red lamps were illuminated during these components. Sessions consisted of 10 FI components separated by 1-min time-out periods during which the stimulus lamps were extinguished and responses had no scheduled consequences. Electromechanical programming and recording equipment was located in a nearby room. Sessions were approximately 40 min and were conducted Monday through Friday.

Drugs

Drug administration was initiated when response rates were stable. Test sessions were conducted on Tuesdays and Fridays providing that response rates were stable on the preceding day. Drugs were injected into the calf or thigh muscle in a 1.0 ml/kg volume. All of the compounds tested were dissolved in 0.9% NaCl. Ipsapirone was obtained from Miles Pharmaceuticals, West Haven, CT; buspirone and BMY 7378 [8-(2-(4-(2-methoxyphenyl)-1-piperazinyl)ethyl)-8-azaspirol(4.5)-decane-7,9-dione dihydrochloride] from Bristol-Myers, Wallingford, CT; 8-OH-DPAT [8-hydroxy-2-(di-*n*-propylamino)tetralin] from Research Biochemicals, Inc., Natick, MA; flesinoxan from Duphar, B.V., Weesp, The Netherlands; RU 24969 [5-methoxy-3(1,2,3,6-tetrahydropyridin-4-yl)1H indole] from Roussel UCLAF, Paris; SM 3997 (3aα,4β,7β,7aα-hexahydro-2-(4-(4-(2-pyrimidinyl)-1-piperazinyl)-butyl-4,7-methano-1*H*-isoindole-1,3(2H)-dione dihydrogen citrate) from Pfizer Pharmaceuticals, New York; WY 47,846 [3a,4,4a,6a,7,7a-hexahydro-2-(4-(4-(2-pyrimidinyl) butyl)-4,7-etheno-1H-cyclobut(f)isoindole-1,3(2H)-dione] from Wyeth-Ayerst, Princeton, NJ; and midazolam from Hoffmann-La Roche, Inc., Nutley, NJ.

Data Analysis

The effects of each drug dose on response rate were calculated as the percent of control response rates determined during that dose-response curve. Control data were taken from Thursday sessions. Drug effects were considered significant in each monkey if they differed by 2 standard deviations or more from that monkey's control rate. The data in Fig. 1 are the averages across all monkeys receiving the drugs. Each drug was tested in four or five monkeys.

RESULTS

Control performance was characterized by low rates and some positive acceleration within each 3-min interval. Average control rate was 0.16 responses/sec (±0.03 s.e.m.); mean response rate prior to the introduction of shock was 0.74 (±0.25 s.e.m.) responses/sec.

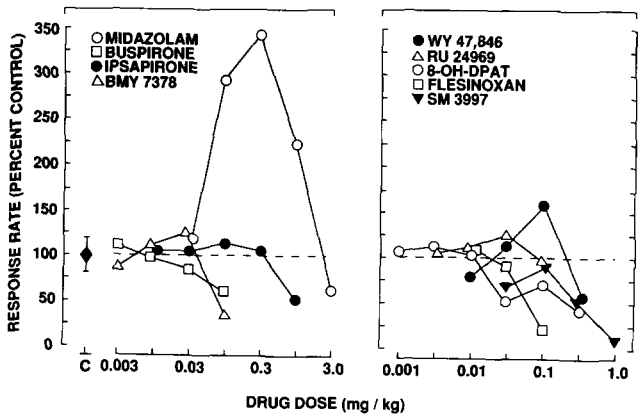

FIG. 1. Effects of midazolam and 5-HT_{1A} agonists on punished responding of squirrel monkeys. The unconnected point on the left represents control responding; vertical bars represent ±1 s.d. The horizontal dashed line indicates 100% control response rate. Percent control response rate was determined for each subject relative to its own control rate, then averaged across subjects to obtain the points depicted in the figure.

Midazolam increased responding substantially. The dose-response curve is presented in the left panel of Fig. 1. Doses of 0.1–1.0 mg/kg resulted in response rates significantly higher than control rates in three or four of the four monkeys tested at each of these doses. The highest dose tested, 3.0 mg/kg, decreased responding below baseline levels in two of three monkeys.

None of the other drugs tested produced effects similar in magnitude to those exhibited by midazolam. Some response rate increases were observed at one or two doses of each of these drugs except flesinoxan and SM 3997, but only in one or two of the monkeys tested with each drug. With the exception of RU 24969, all of these drugs decreased response rates significantly at one or more of the higher doses.

DISCUSSION

The large increases in responding produced by midazolam and the lack of effect observed with the 5-HT_{1A} agonists are similar to results obtained in conflict procedures in rats, but differ from results reported in pigeons. The effects of midazolam are consistent with other reports in monkeys that benzodiazepines produce large increases in punished responding. Buspirone has been reported to increase punished responding of both cynomolgus and squirrel monkeys (9,24), but the present results are consistent with a study by Wettstein (25), using similar operant procedures, in which buspirone either had no effect or decreased responding in squirrel monkeys. The present results also are not entirely incompatible with the Weissman *et al.* (24) study, in which buspirone's effects were much smaller in magnitude than those of midazolam, and occurred under a different baseline schedule of reinforcement that produced very low control rates of responding. In contrast to the fixed-interval schedule of food delivery with fixed-ratio shock used in the present experiment and by Wettstein, Weismann *et al.* also used a fixed-ratio food schedule with superimposed fixed-ratio shock. This schedule engendered much lower response rates, and as a result, greater increases in response rates expressed as percent control.

The lack of effect of buspirone is not unique to this drug, which has other effects besides acting as a 5-HT_{1A} agonist. Neither ipsapirone nor BMY 7378, structural analogs of buspirone without

its dopamine antagonist properties (5,26), produced increases in responding, nor did RU 24969, a mixed 5-$HT_{1A/1B}$ agonist that exerts substantial 5-HT_{1B}-mediated effects in rats (12). Flesinoxan, SM 3997, WY 47,846, and 8-OH-DPAT are probably more selective for the 5-HT_{1A} site than the other drugs, but none of these compounds increased responding.

Autoradiographic studies have shown some differences in the distribution of 5-HT_1 binding sites in pigeons, rats, and monkeys (23). Rats and mice appear to be unique in having 5-HT_{1B} binding sites; this site has not been identified in either pigeon or monkey brain. Waeber *et al.* (23) suggest that the majority of sites in the pigeon brain are of the 5-HT_{1D} subtype. In the monkey brain, 5-HT_{1A} binding sites are numerous in the neocortex, hippocampus, and dorsal raphe nucleus, as they are in rats. Thus, the species differences observed in the anticonflict effects of 5-HT_{1A} agonists may reflect a differential anatomical distribution and/or functional role for this binding site in pigeons as opposed to mammals.

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