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Short communication

IN VIVO EVIDENCE OF PARTIAL AGONIST ACTIVITY EXERTED BY PURPORTED 5-HYDROXYTRYPTAMINE ANTAGONISTS

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Using a food-reinforced two-lever operant method, rats (n = 9) were trained to discriminate 0.16 mg/kg LSD from saline. Tests for stimulus generalization in rats so trained indicated that the purported 5-HT antagonists cyproheptadine (1.25 and 10 mg/kg), methysergide (0.16 to 10 mg/kg) and mianserin (2.5 to 40 mg/kg) produced partial generlization with LSD. The hallucinogens mescaline (5 to 40 mg/kg) and quipazine (1.25 to 5 mg/kg) were also generalized with LSD. The data suggest that cyproheptadine, methysergide and mianserin may produce partial agonist effects in addition to their antagonist action at central 5-HT receptor sites.

Cyproheptadine | Drug discrimination | 5-HT agonist/antagonist
LSD cue | Methysergide | Mianserin

1. Introduction

A number of purported antagonists of 5-hydroxytryptamine (5-HT) are currently being used as pharmacological tools in studying the role of 5-HT in the central nervous system, and in elucidating the mechanism of action of other centrally acting drugs (Garattini and Samanin, 1978). In support of their presumed 5-HT blocking activity, compounds such as methysergide and cyproheptadine have been shown (Appel et al., 1978; Kuhn et al., 1978; Winter, 1978) to antagonize the discriminative stimulus properties of lysergic acid diethylamide (LSD), a hallucinogen which may act by mimicking 5-HT at central 5-HT receptor sites (Aghajanian, 1972). However, preliminary studies in this laboratory suggested that several purported 5-HT antagonists may exert agonist effects in addition to their known antagonist activity.

The present study was aimed at determining whether the purported 5-HT receptor blockers cyproheptadine, methysergide and mianserin (Gyermek, 1966; Garattini and Samanin, 1978) may possess in vivo agonist effects in the rat. The drug discrimination test was used for this purpose because it has been found (Colpaert et al., 1976; Holtzman et al., 1977) capable of characterizing the agonist effects of partial agonist/antagonists of the narcotic class of compounds.

2. Materials and methods

The drug discrimination procedure used here has been described in much detail elsewhere (Colpaert et al., 1976). In short, materials consist of standard animal test cages, fitted with 2 levers and a food cup, and programmed by solid-state logic modules. 9 rats were trained to bar press for food (45-mg pellets) on either of 2 levers; the contingency was such that one pellet was delivered after every 10th response (FR 10). Following i.p. injection of 0.16 mg/kg LSD tartrate 15 min before the session, the animals were required

to press one of the levers (drug lever) in order to get reinforcement; on i.p. saline injection, the rats were required to press the opposite lever (saline lever). Every week, each rat was run in daily 15-min sessions on 5 consecutive days. LSD or saline injections were given according to alternating sequences (Colpaert et al., 1976). Lever selection on every single session was considered to be correct if the animal made fewer than 10 responses on the inappropriate lever, before completing 10 responses on the appropriate one. The total number of responses was also recorded after each session. The discrimination training was continued for at least 3 months; further training was given to those animals which had not reached a criterion consisting of 10 consecutive sessions on which lever selection was correct. Following training, test sessions were held on Wednesdays and Fridays; on the remaining 3 days, the training conditions were continued to ensure reliable performance. In test sessions, the rats were treated with the injection conditions being studied, and put into the test cage after the required time interval. The animals then were to select one of the 2 levers. That is, the lever on which the rat totalled 10 responses first was regarded as the selected lever; subsequent reinforcement was made contingent upon pressing (FR 10) the selected lever.

1 h following any (training/test) session, the animals were allowed to feed freely for 2 h; tap water was continuously available in the individual home cages.

The antagonists being studied were: 1.25, 2.5, 5 and 10 mg/kg cyproheptadine HCl; 0.16, 0.63, 2.5 and 10 mg/kg methysergide maleate; and 2.5, 10 and 40 mg/kg mianserin HCl. The antagonists were injected subcutaneously, 60 min before the test, because the present results were also to serve as basis for further antagonist studies. As an additional control, the animals were also given test sessions on each of 3 subcutaneous saline injections; they invariably selected the saline lever, and this part of the data is not reported in the Results section.

To substantiate the agonist specificity of the LSD cue (Appel et al., 1978) under the present conditions, generalization tests were also conducted on 5 to 40 mg/kg mescaline HCl, and on 1.25 to 5 mg/kg quipazine maleate. These two drugs were injected i.p. 15 min before the test.

Randomly selected subgroups of rats were tested on several doses of each of the drugs being studied. Cyproheptadine, methysergide, mianserin and quipazine were studied in 5 rats each; 6 rats were used for tests with mescaline. The sequence of testing was randomized for each rat individually. Doses of test drugs were selected on the basis of preliminary data obtained in 3 rats trained to discriminate 0.16 mg/kg LSD from saline; these data are not included in the present report. The injection volume was 1 ml/100 g body weight.

3. Results

The data are summarized in fig. 1. The hallucinogens mescaline (5 to 40 mg/kg) and quipazine (1.25 to 5 mg/kg) produced a dose-dependent increase in the number of rats selecting the drug lever, thus indicating that these drugs produce stimulus generalization with the training drug, LSD. The stimulus generalization gradient of these two compounds was relatively steep, and the effect reached the 100% level with the 40 and 5 mg/kg doses, respectively. Both compounds also exerted an almost linear rate-decreasing effect, and 5 mg/kg quipazine depressed the response rate by nearly 60%.

Within the 1.25 to 5 mg/kg dose-range, cyproheptadine also produced a dose-dependent stimulus generalization; at 10 mg/kg, however, the incidence of generalization declined rather than increased. Unlike its generalization curve, the rate-depressant effects of cyproheptadine were proportional to the dose throughout the entire 1.25 to 10 mg/kg range; the depressant effect amounted to as much as 80% with the 10 mg/kg dose. The

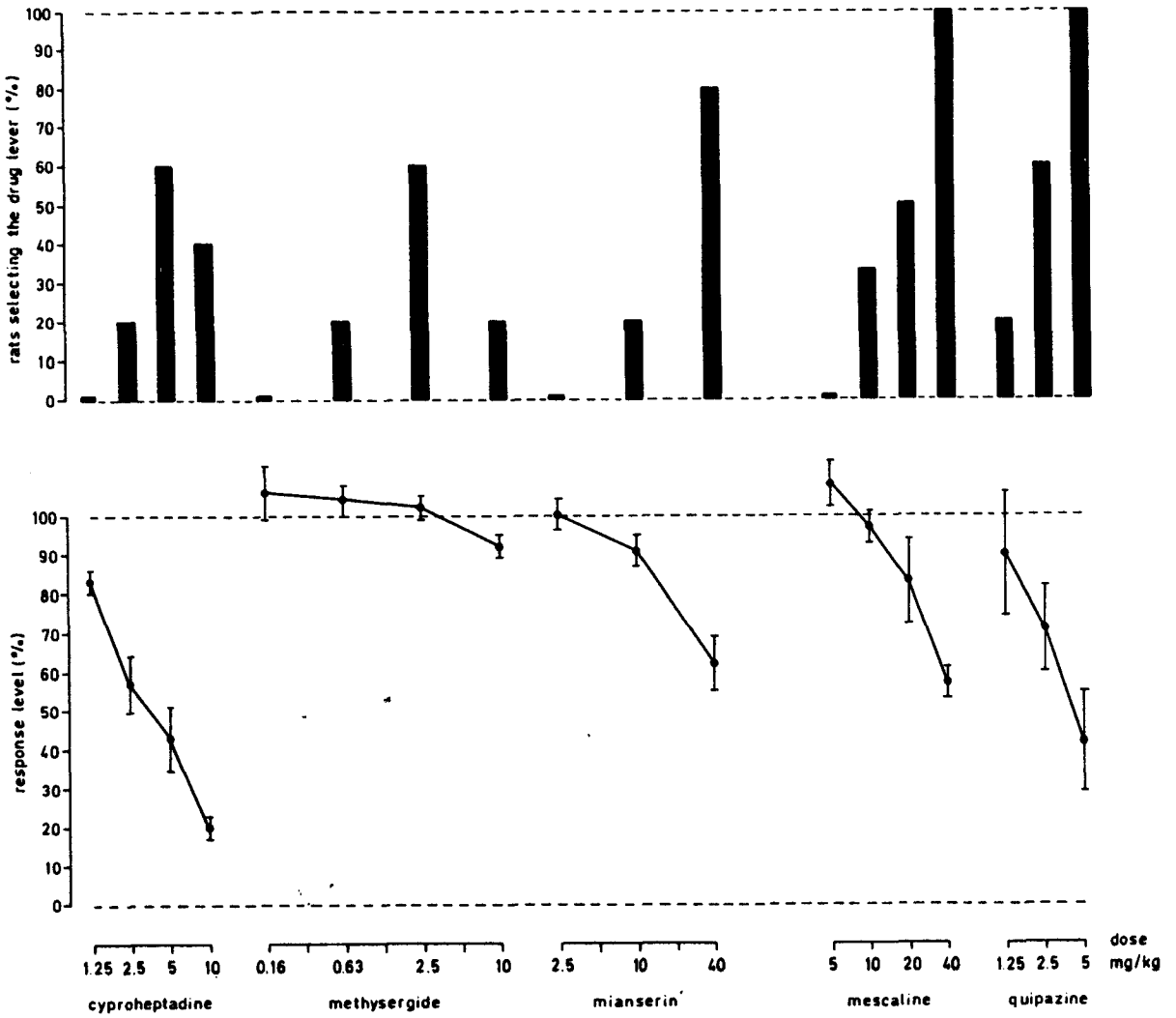

Fig. 1. Stimulus generalization of 5-HT antagonists and agonists in rats trained to discriminate 0.16 mg/kg LSD from saline. Upper ordinate: percent of rats selecting the drug lever; the animals were trained to select the drug lever on injection of 0.16 mg/kg LSD, and to select the saline lever on saline injection. Response level indicates the total number of responses as a percent of the number of responses on the most recently preceding saline control sessions (Colpaert et al., 1976). Response level is expressed as the mean ± 1 S.E.M. Cyproheptadine, methysergide, mianserin and quipazine were tested in 5 rats each; mescaline was tested in 6 rats.

stimulus generalization which occurred with 0.16 to 10 mg/kg methysergide was similar in shape to that of cyproheptadine, as was the maximum intensity of effect (60%). In contrast with cyproheptadine, however, methysergide failed to exert a marked rate-depressant effect at any of the doses tested. Mianserin induced a stimulus generalization which was directly proportional to the dose within the 2.5 to 40 mg/kg range; 160 mg/kg was not tested because of the apparent ill effects of the drug at this dose in experimentally naive rats. The doses 10 and 40 mg/kg of mianserin were associated with rate-

depressant effects, amounting to about 40% depression at the highest dose tested.

4. Discussion

The data obtained in this study (fig. 1) indicate that the purported 5-HT antagonists (Gyermek, 1966; Garattini and Samanin, 1978) cyproheptadine, methysergide and mianserin mimick the discriminative stimulus properties of LSD in rats trained to discriminate 0.16 mg/kg LSD from saline. This finding substantiates the hypothesis that these compounds exert intrinsic agonist effects in addition to their known 5-HT receptor blocking action. The generalization which occurred with cyproheptadine and methysergide was characterized by two distinctive features. Firstly, in both cases the generalization was partial in that it failed to reach the 100% level of effect. Secondly, the generalization was not directly proportional to dose over the entire dose-range, and it appears that the data can best be approximated by an inverted U-shaped curve. These two features of generalization data are known (Colpaert et al., 1976; Holtzman et al., 1977) to be characteristic of the effects of partial narcotic agonists/antagonists such as cyclazocine and nalorphine in animals trained to discriminate a pure narcotic agonist from saline. Unlike cyproheptadine and methysergide, mianserin produced a monophasic dose-response curve and a higher maximum effect; a similar generalization occurs (Colpaert et al., 1976) with partial narcotic agonists/antagonists whose agonist activity is relatively more prominent (e.g. pentazocine). The present data therefore suggest that cyproheptadine, methysergide and mianserin exert partial agonist/antagonist activity, and may hence not be considered as pure antagonists. The agonist activity revealed here is even more important as it occurred in rats trained on what appears to be a relatively high training dose (Appel et al., 1978) of LSD. According to related drug discrimination studies (Shannon and Holtzman, 1979), it is reasonable to expect that an even higher incidence of generalization might occur in rats trained on a lower LSD dose.

The present data confirm earlier evidence that 5-HT agonist (Aghajanian, 1972; Garattini and Samanin, 1978) and hallucinogenic drugs such as mescaline and quipazine, produce stimulus generalization with LSD. With both drugs, the generalization was directly proportional to the dose, and reached the 100% effect level (fig. 1). The discriminative stimulus properties of mescaline (Browne and Ho, 1975) and quipazine (White et al., 1979; Winter, 1979) like those of LSD itself (Appel et al., 1978; Kuhn et al., 1978; Winter, 1978), are likely to be associated with their 5-HT agonist action. This further supports that the effects observed here with the purported antagonists cyproheptadine, methysergide and mianserin are contingent on a partial agonist action of these drugs at 5-HT receptor sites in the central nervous system.

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