Pharmacology Biochemistry and Behavior, Vol. 42, pp. 519–522, 1992
Printed in the U.S.A.

0091-3057/92 $5.00 + .00

RAPID COMMUNICATION

MDL 73,147EF, a 5-HT_3 Antagonist, Facilitates Latent Inhibition in the Rat

PAULA M. MORAN[1] AND PAUL C. MOSER

Marion Merrell Dow Research Institute, 16 rue d'Ankara, Strasbourg, France

Received 27 January 1992

MORAN, P. M. AND P. C. MOSER. *MDL 73,147EF, a 5-HT_3 antagonist, facilitates latent inhibition in the rat.* PHARMACOL BIOCHEM BEHAV **42**(3) 519–522, 1992.—Latent inhibition (LI) is a behavioral model of selective attention that has been used to study the attentional deficits seen in schizophrenia. In the present study, we examined the effect of 5-hydroxytryptamine$_3$ (5-HT_3) receptor blockade on LI using the conditioned emotional response (CER) procedure. Prior exposure to 20, 30, or 40 stimulus presentations significantly, and almost completely, inhibited the CER to that stimulus. This LI effect was much weaker when only 10 preexposures were given. 1*H*-indole-3-carboxylic acid, trans-octahydro-3-oxo-2,6-methano-2*H*-quinolizin-8-yl ester methanesulfonate (MDL 73,147EF), a selective 5-HT_3 receptor antagonist, significantly facilitated the LI effect observed after 10 preexposures at 0.1 mg/kg but not at 0.01 mg/kg. The magnitude of this effect was comparable to that observed with the classical neuroleptic haloperidol (0.1 mg/kg). Neither MDL 73,147EF nor haloperidol affected the CER in animals not preexposed to the stimulus. These results strongly corroborate suggestions that 5-HT_3 receptor antagonists will be of use in the treatment of schizophrenia.

Latent inhibition Schizophrenia MDL 73,147EF 5-HT_3 receptors

A considerable amount of recent data supports the hypothesis that 5-hydroxytryptamine$_3$ (5-HT_3) receptor antagonists can modulate activity of the mesolimbic dopamine system [for review, see (4)]. On the basis of these findings, it has been suggested that 5-HT_3 receptor antagonists might represent a novel approach to antipsychotic therapy (4). However, the exact role of dopamine in the pathology of schizophrenia is far from clear (14) and the approach used to assess the potential antipsychotic activity of 5-HT_3 receptor antagonists, in which specific dopamine systems are artificially activated, makes implicit assumptions about such a role. An alternative approach is to examine the effects of these compounds in animal models relevant to the behavioral deficits observed in schizophrenics. One such model is latent inhibition (LI) [for review, see (5)]. LI refers to a process of learning to ignore irrelevant stimuli and is manifested behaviorally in both animals and humans by the finding that repeated exposure to a nonreinforced stimulus retards subsequent conditioning to that stimulus. During an acute phase of their disorder, schizophrenics show a deficit in LI whereas no such deficit is seen in chronic schizophrenics (2). In rats, LI is enhanced by administration of both classical and atypical neuroleptics (3,5,6).

In the following study, we examined the effect of 1*H*-indole-3-carboxylic acid, trans-octahyro-3-oxo-2,6-methano-2*H*-quinolizin-8-yl ester methanesulfonate (MDL 73,147EF), a novel 5-HT_3 receptor antagonist (7), on LI in rats using the conditioned emotional response (CER) procedure described by Weiner et al. (20). Essentially, the procedure consists of four stages: a) habituation of rats to drink from a water spout immediately on being placed in the experimental chamber; b) preexposure to the to-be-conditioned stimulus (tone); c) conditioning, during which the tone stimulus is paired with mild foot-shock; and d) testing, when the degree of suppression of drinking elicited by the tone stimulus is measured. Under these conditions, LI is observed as a decrease in this suppression in animals preexposed to the tone compared to those not preexposed to it. The strength of the LI effect covaries with the amount of preexposure and in the present experiments we used a low number of stimulus preexposures to evaluate possible potentiating effects of MDL 73,147EF.

METHOD

Animals

Male Sprague-Dawley rats (Charles River, France) weighing 260–320 g were used for all experiments. They were housed

[1] Requests for reprints should be addressed to Paula M. Moran, Marion Merrell Dow Research Institute, 16 rue d'Ankara, B.P. 447 R/9, F-67009 Strasbourg, Cédex, France.

five per cage in a temperature-controlled (20 ± 2°C) environment on a 12 L:12 D schedule. Animals were placed on a 23.5-h water deprivation schedule 48 h prior to, and throughout, testing. Food was freely available in the home cage.

Apparatus

All experimental procedures were carried out in a dark Perspex box (20 × 20 × 20 cm) located within a sound-attenuating chamber containing a loudspeaker. A ventilation fan provided stable background noise. A removable drinking spout was located on one wall of the box. Licks at this spout were counted using a lickometer (Anxiometer model 102; Columbus Instruments, Columbus, OH) connected to a computer system (MED-PC system; Med Associates Inc., East Fairfield, VT) that timed experimental events and also controlled presentation of tone stimuli (400 Hz, 10 s; audio generator LI 100-22; Letica Instruments, Barcelona, Spain) and shock (0.8 mA, 3 s; shock generator and scrambler unit LI 100-20; Letica Instruments, Spain). Scrambled foot-shock was applied via the floor of the Perspex box, which consisted of metal bars (2.5 mm diameter) spaced 14 mm apart.

Procedure

The experimental procedure used, essentially that of Weiner et al. (20), can be divided into four phases:

1. *Habituation* (days 1-6). Animals were placed in the experimental chamber and remained there until they had completed 1,000 licks or 10 min had elapsed. Any animals that did not complete 1,000 licks within 10 min on any of the last 3 days of habituation were excluded from the experiment.
2. *Preexposure* (day 7). Animals were replaced in the experimental chamber with the water spout removed and either 0,10,20,30, or 40 tone presentations were given within a 15-min session.
3. *Conditioning* (day 8). Animals were replaced in the experimental chamber with the water spout removed. Five minutes later, animals were given two tone presentations, 5 min apart. Each tone presentation was immediately followed by scrambled foot-shock (3 s, 0.8 mA). Five minutes after the second tone-shock presentation, animals were removed.
4. *Testing* (day 9). The water spout was replaced in the experimental chamber and animals allowed to drink freely until 100 licks had been made. There then followed a tone presentation that continued until either the animal made a further 10 licks or 300 s had elapsed. A suppression ratio (SR) was calculated as the time between licks 90-100 divided by the time between licks 90-110 (or between lick 90 and the end of the 300-s cutoff period). Animals that were not preexposed to the tone were included to control for possible effects of drug treatment of the CER itself.

TABLE 1

EFFECT OF INCREASING THE NUMBER OF STIMULUS PREEXPOSURES ON SR IN UNTREATED RATS

Number of preexposures	*n*	SR Median (interquartile range)
0	10	0.02 (0.01-0.06)
10	10	0.06 (0.01-0.48)
20	8	0.49 (0.24-0.54)*
30	10	0.50 (0.34-0.52)†
40	10	0.42 (0.01-0.52)*

*$p<0.05$, †$p<0.01$ compared to 0 preexposure group (two-tailed Gehan's test).

Drug Administration

Drugs were dissolved in saline (0.9%) and administered SC prior to preexposure (day 7) and conditioning (day 8) only. Haloperidol (0.1 mg/kg, prepared from commercially available 5-mg/ml HALDOL vials; Laboratoires Janssen, Boulogne-Billancourt, France) was given 60 min before testing; MDL 73,147EF (0.01, 0.1 mg/kg) was given 30 min prior to testing. A dose volume of 1 ml/kg was used.

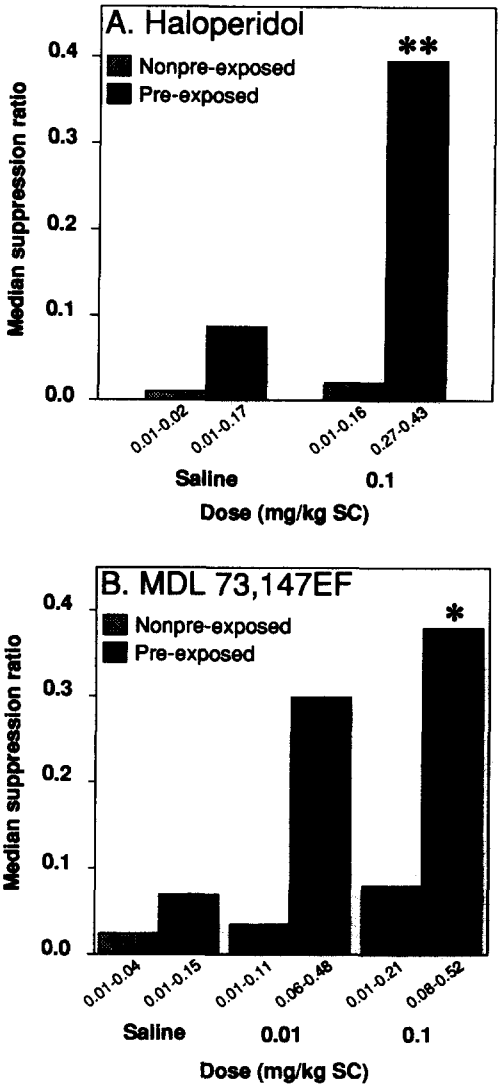

FIG. 1. Panels A and B demonstrate the potentiation of SR by haloperidol and MDL 73,147EF, respectively, in rats that received either 0 or 10 tone preexposures. *$p < 0.05$, **$p < 0.01$ compared to preexposed saline group (Gehan's test). The values shown are the medians from 10 rats. The values below each column are the interquartile ranges.

Statistics

As a cutoff was used in calculating SR, statistical comparisons between groups were carried out using a two-tailed Gehan's test for censored data (11).

RESULTS

Increasing the number of tone preexposures from 0 to 40 resulted in a marked increase in SR (Table 1). In the 0-exposure condition, animals showed the expected CER during the test phase of the experiment, that is, drinking was effectively suppressed as indicated by the low SR. Following preexposure to 20, 30, or 40 tone presentations, the SR was significantly and almost maximally increased, demonstrating a robust LI effect. Following 10 preexposures, SR values were only slightly greater than those of non-preexposed animals; this condition was therefore used for the subsequent analysis of drug effects so that increases in SR, indicative of a facilitation of LI, could be detected.

As previously reported (3,6,19), pretreatment with haloperidol (0.1 mg/kg) significantly potentiated the effect of 10 tone preexposures on SR (Fig. 1A). A similar effect was observed following administration of 0.1 mg/kg MDL 73,147EF (Fig. 1B). At the lower dose of 0.01 mg/kg, an increase in LI was observed, although this effect failed to reach statistical significance ($p = 0.058$). Neither haloperidol nor MDL 73,147EF, at the doses tested in the present study, had any effect on SR in animals not preexposed to the tone, demonstrating that they did not disrupt the CER itself (Fig. 1).

DISCUSSION

Increasing the number of tone preexposures from 0 to 40 significantly increased SR, in agreement with a number of previous studies (3,19). This relationship between number of preexposures and SR showed that LI could be demonstrated under these experimental conditions and allowed us to select a preexposure condition that would enable a facilitation of LI to be detected. Haloperidol (0.1 mg/kg) significantly potentiated the low SR value seen following 10 preexposures, confirming the facilitatory effect of this drug on LI (3,19). MDL 73,147EF, a selective 5-HT_3 receptor antagonist (7,17), also significantly increased the SR under these conditions. This increase was of a magnitude comparable to that seen with haloperidol and, like the effect of haloperidol, was observed in the absence of any significant effects on the CER. Facilitation of LI has previously been demonstrated for a range of both typical and atypical neuroleptic agents, suggesting that such an effect following the 5-HT_3 receptor antagonist MDL 73,147EF may be predictive of antipsychotic activity (5,6).

In addition to their possible antipsychotic potential, 5-HT_3 receptor antagonists have been shown to have anxiolytic and cognition-enhancing activity in animals (1,4). Although the CER can be used as an animal model of anxiety (18), a possible anxiolytic action of MDL 73,147EF in mediating its facilitation of LI is unlikely: First, no drug was administered on the test day and, second, administration of drug on the 2 days prior to testing had no effect on the CER in the absence of stimulus preexposure. 5-HT_3 receptor antagonists have also been reported to enhance cognition in both rodents and primates (1). As LI is considered to involve selective attentional processes (8,12), its facilitation by a 5-HT_3 receptor antagonist suggests that the reported promnesic effects of these compounds may be consequent to, or at least be in part attributable to, enhancement of selective attention. However, this possibility would have to be tested in a more direct comparison of promnesic and LI facilitatory effects of 5-HT_3 receptor antagonists.

At present, the exact mechanism of action of MDL 73,147EF in facilitating LI is unknown. A number of studies have demonstrated that 5-HT_3 receptor antagonists, including MDL 73,147EF, can suppress increased activity of dopaminergic neurones within the mesolimbic system, particularly that part projecting to the nucleus accumbens (4,13,17). This system has also been implicated in LI, which is disrupted by injections of amphetamine into the nucleus accumbens (16). However, the involvement of other brain areas cannot yet be ruled out as, in addition to the nucleus accumbens, several other brain areas that have a high density of 5-HT_3 receptors, such as the amygdala and hippocampus (10), have also been implicated in LI (9,15).

In conclusion, the present study strongly corroborates suggestions that 5-HT_3 receptor antagonists may have antipsychotic potential (4). Previously, this proposition was based upon studies of the interactions of 5-HT_3 receptor antagonists with CNS dopaminergic systems. Our results extend those findings to include activity of a 5-HT_3 receptor antagonist in a behavioral model relevant to the information-processing deficits seen in schizophrenia (5,8).

ACKNOWLEDGEMENT

The authors thank Thierry Fischer for excellent technical assistance.

REFERENCES

1. Barnes, J. M.; Costall, B.; Coughlan, J.; Domeny, A. M.; Gerrard, P. A.; Kelly, M. E.; Naylor, R. J.; Onaivi, E. S.; Tomkins, D. M.; Tyers, M. B. The effects of ondansetron, a 5-HT_3 receptor antagonist, on cognition in rodents and primates. Pharmacol. Biochem. Behav. 35:955–962; 1990.
2. Baruch, I.; Hemsley, D. R.; Gray, J. A. Differential performance of acute and chronic schizophrenics in a latent inhibition task. J. Nerv. Ment. Dis. 176:598–606; 1988.
3. Christison, G. W.; Atwater, G. E.; Dunn, L. A.; Kilts, C. D. Haloperidol enhancement of latent inhibition: Relation to therapeutic action? Biol. Psychiatry 23:746–749; 1988.
4. Costall, B.; Naylor, R. J.; Tyers, M. B.; The psychopharmacology of 5-HT_3 receptors. Pharmacol. Ther. 47:181–202; 1990.
5. Ellenbroek, B. A.; Cools, A. R. Animal models with construct validity for schizophrenia. Behav. Pharmacol. 1:469–490; 1990.
6. Feldon, J.; Weiner, I. The latent inhibition model of schizophrenic attention disorder: Haloperidol and sulpiride enhance rats' ability to ignore irrelevant stimuli. Biol. Psychiatry 29:635–646; 1991.
7. Gittos, M. W.; Fatmi, M. Potent 5-HT_3 antagonists incorporating a novel bridged pseudopelletierine ring system. Actual. Chim. Thér. 16:187–198; 1989.
8. Gray, J. A.; Feldon, J.; Rawlins, J. N. P.; Hemsley, D. R.; Smith, A. D. The neuropsychology of schizophrenia. Behav. Brain Sci. 14:1–84; 1991.
9. Kaye, H.; Pearce, J. M. Hippocampal lesions attenuate latent inhibition of a CS and of a neutral stimulus. Psychobiology 15: 293–299; 1987.
10. Kilpatrick, G. J.; Jones, B. J.; Tyers, M. B. Identification and distribution of 5-HT_3 receptors in rat brain using radioligand binding. Nature 330:746–748; 1987.

11. Krauth, J. Distribution-free statistics: An application-oriented approach. Amsterdam: Elsevier; 1988.
12. Mackintosh, N. J. A theory of attention: Variations in the associability of stimuli with reinforcement. Psychol. Rev. 82:276–298; 1975.
13. Moser, P. C. Effect of chronic treatment with 5-HT_3 receptor antagonists on monoamine concentrations in various regions of the rat brain. Br. J. Pharmacol. 102:143P; 1991.
14. Reynolds, G. P. Beyond the dopamine hypothesis: The neurochemical pathology of schizophrenia. Br. J. Psychiatry 155:305–316; 1989.
15. Salafia, W. R.; Allan, A. M. Augmentation of latent inhibition by electrical stimulation of hippocampus. Physiol. Behav. 29: 1125–1130; 1982.
16. Solomon, P.; Statton, D. Differential effects of microinjections of *d*-amphetamine into the nucleus accumbens or the caudate putamen on the rat's ability to ignore an irrelevant stimulus. Biol. Psychiatry 17:743–756; 1982.
17. Sorensen, S. M.; Humphreys, T. M.; Palfreyman, M. G. Effect of acute and chronic MDL 73,147EF, a 5-HT_3 receptor antagonist, on A9 and A10 dopamine neurones. Eur. J. Pharmacol. 163:115–118; 1989.
18. Treit, D. Animal models for the study of anti-anxiety agents: A review. Neurosci. Biobehav. Rev. 9:203–222; 1985.
19. Weiner, I.; Feldon, J. Facilitation of latent inhibition by haloperidol in rats. Psychopharmacology 91:248–253; 1987.
20. Weiner, I.; Lubow, R. E.; Feldon, J. Disruption of latent inhibition by acute administration of low doses of amphetamine. Pharmacol. Biochem. Behav. 30:871–878; 1988.