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Modulation of phencyclidine-induced changes in locomotor activity and patterns in rats by serotonin

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Abstract

To test the hypothesis that serotonergic modulation of the effects of phencyclidine (PCP) are due to circuit- rather than receptor-based interactions between glutamatergic and serotonergic systems, multivariate profiles of rat behavior were assessed after treatments with the 5-hydroxytryptamine (5-HT) 5-HT_2 receptor antagonist ketanserin (1.0 mg/kg), the 5-HT_2 receptor agonist (1(2,5-dimethoxy-4-iodophenyl)-2-aminopropane) (DOI; 0.27 mg/kg), various doses of PCP (0.75 to 10.125 mg/kg), or combinations thereof. Ketanserin blocked all effects of DOI, but reduced the effects of PCP only on locomotion. Depending on the dose, PCP was observed to increase or decrease locomotion and the roughness of the rats' patterns of locomotion. In any case, DOI always increased the activity and decreased the roughness of locomotor paths in PCP-treated rats. Thus, co-administration of DOI and PCP did not yield a shift in the dose–effect curve for either drug, but instead resulted in a new behavioral profile consistent with a circuit-based dynamic interaction. © 1998 Elsevier Science B.V.

Keywords: Phencyclidine; DOI (1(2,5-dimethoxy-4-iodophenyl)-2-aminopropane); Ketanserin; Locomotion; 5-HT_2 receptor; (Rat)

1. Introduction

Phencyclidine (PCP) is a drug of abuse that has a wide range of psychotomimetic effects in humans (Luby et al., 1959, 1962). Many researchers have drawn parallels between these effects of PCP in humans and some symptoms of schizophrenia (Luby et al., 1959, 1962; Garey, 1979; Domino and Luby, 1981; Snyder, 1988; Javitt and Zukin, 1991a,b). While not a perfect model of schizophrenia, PCP has been utilized for developing animal models of the effects of antipsychotic drugs. Accordingly, many compounds have been tested for their ability to alter the effects of PCP in animals. These experiments are of interest from a practical, clinical standpoint, but also from a theoretical one, as drugs which alter the expression of PCP-induced behavior may be informative about how PCP works to produce these effects (Murray and Horita, 1979; Castellani and Adams, 1981; Yamaguchi et al., 1986; Lehmann-Masten and Geyer, 1991; Jackson et al., 1994; Kitaichi et al., 1994; Ögren and Goldstein, 1994; Carlsson, 1995; Corbett et al., 1995; Lapin and Rogawski, 1995; Maurel-Remy et al., 1995; Svensson et al., 1995; Tsutsumi et al., 1995; Gleason and Shannon, 1997).

PCP is a noncompetitive antagonist at NMDA receptors which also acts as an indirect releaser of dopamine and serotonin (5-hydroxytryptamine, 5-HT) and as a monoamine reuptake inhibitor (Garey and Heath, 1976; Smith et al., 1977; Hernandez et al., 1988; Kameyama et al., 1988; Rao et al., 1989; Johnson and Jones, 1990; Steinpreis and Salamone, 1993; Ali et al., 1994; Lillrank et al., 1994; Hertel et al., 1996; Hori et al., 1996). PCP produces a variety of unusual behaviors in rodents, dominated by increases in locomotor activity and stereotyped behaviors.

These PCP-induced effects on locomotor activity and stereotyped behaviors have been altered by various pharmacological manipulations. Many researchers have examined the influences of serotonergic manipulations, particularly 5-HT_2 receptor antagonists, on PCP-induced locomotor or stereotyped behavior. The ability of 5-HT_2 receptor antagonists to alter the effects of PCP, however, varies with the behavioral measures examined. Pretreatment with

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5-HT_2 or 5-HT_{2A} receptor antagonists has been reported to block the hyperactivity or stereotyped behaviors produced by PCP (Nabeshima et al., 1987a,b; Yamaguchi et al., 1987; Kitaichi et al., 1994; Maurel-Remy et al., 1995; Gleason and Shannon, 1997), while other researchers have failed to see this antagonism (Nabeshima et al., 1987a,b; Yamaguchi et al., 1987; Jackson et al., 1994; Kitaichi et al., 1994). The importance of 5-HT_2 receptors in the effects of PCP-induced behavior is emphasized by the observations that PCP-induced hyperlocomotion in rats and mice was also blocked by the atypical antipsychotic clozapine and the putative atypical antipsychotics olanzapine and risperidone and at doses lower than those which reduced activity by themselves. These antipsychotics, while active at numerous sites, are also known to be 5-HT_2 receptor antagonists. Indeed, the ability to block PCP correlated highly with the ability to antagonize 5-HT_2 receptors (Maurel-Remy et al., 1995; Gleason and Shannon, 1997). Hence, 5-HT_2 receptor antagonists block some effects of PCP, but not others.

The fact that antagonism of 5-HT_2 receptors attenuates only some behavioral effects of PCP is inconsistent with an interaction between serotonin and PCP at a particular receptor or even within a particular class of synapses. If the effects of PCP on locomotor activity are influenced by 5-HT_2 receptor modulation through a circuit interaction involving multiple brain regions, a functional interaction between 5-HT_2 receptor modulation and PCP should be demonstrable with either a 5-HT_2 receptor antagonist, as seen above, or a 5-HT_2 receptor agonist. In fact, a 5-HT_2 receptor agonist, because it is active, might actually be more likely to demonstrate such an interaction than a silent antagonist. Hence, we examined the effects of both a 5-HT_2 receptor antagonist and a 5-HT_2 receptor agonist on the effects of PCP on multiple measures of locomotor and investigatory behavior.

To systematically test the influence of 5-HT_2 receptors on the effects of PCP, we administered PCP to rats and tested them in our Behavioral Pattern Monitor (BPM), which is an activity and holeboard chamber that enables analyses of quantitative and qualitative changes in patterns of locomotor and investigatory activity (Geyer, 1990). Previous experiments have demonstrated that PCP increased locomotor activity measures in the BPM (Lehmann-Masten and Geyer, 1991). In the current experiments, we explored the effects of PCP on the amount of locomotor activity and the spatial patterns of activity, as well as investigatory behavior. Based on previous evidence that 5-HT_2 receptor antagonists reduce the locomotor-activating effects of PCP, we predicted that a 5-HT_2 receptor agonist would increase and a 5-HT_2 receptor antagonist would decrease the effects of PCP. First, a dose of the 5-HT_2 receptor antagonist ketanserin that would block the effects of the 5-HT_2 receptor agonist DOI (1(2,5-dimethoxy-4-iodophenyl)-2-aminopropane) completely was determined. Second, the influence of this dose of ketanserin on the behavioral effects of a selected dose of PCP was determined. Third, a wide range of doses of PCP was tested with and without a pretreatment of DOI. As far as we know, this is the first time that a 5-HT_2 receptor agonist has been examined for its influences on PCP-induced behavior.

2. Materials and methods

2.1. Animals

Naive male Sprague–Dawley rats (Harlan Industries, San Diego, CA) were housed two or three per cage under a 12 h reverse light cycle (lights off: 07.00 h). Food and water were available ad libitum. Weights ranged from 250 to 350 g. Animals were allowed to acclimatize for approximately 2 weeks after arrival.

2.2. Behavioral testing

All behavior was measured in the BPM, a 30.5×61.0 cm black Plexiglas chamber. Ten 2.5 cm holes are placed in the chamber (three in each long wall, one in one short wall, and three in the floor). Photocells in each hole detect investigatory nosepokes (holepokes). A touchplate, 15.2 cm above the floor, allows detection of rearings when contact is made by the animal between the metal floor and the metal touchplate. A 4×8 array of photobeams is used to define the animal's position in an X–Y coordinate system with a resolution of 3.8 cm. Chambers are kept dark with the exception of 7.5 W red lights. An IBMPC-compatible computer monitors the status of all photobeams and records the duration and nature of all changes with a temporal resolution of 55 ms and stores the data for later analysis.

2.3. Analysis

The raw data were reduced to: the X and Y coordinates of the rat in the chamber, the occurrence of holepokes or rearings, and the amount of time spent at a particular coordinate or performing a particular behavior. Further analyses produced specific measures of behavior. Locomotor activity was quantified by the number of crossings between any of eight equal square sectors within the BPM (crossings) as a measure of horizontal locomotion. Analysis of the spatial structure of rat locomotor paths was assessed by calculating the descriptive statistic, spatial d. Spatial d is based on fractal geometry and is derived using scaling arguments, as described in detail in Geyer et al. (1986) and Paulus and Geyer (1991). Spatial d is calculated by examining the path length through the BPM using several different spatial resolutions. The rate at which the

path length decreases as spatial resolution decreases is fitted to an exponential function for which *d* is a coefficient of the spatial resolution. An increase in *d* reflects an animal's tendency towards rougher, varied, random movements in the path, where the measured path length decreases quickly with decreasing spatial resolution. A decrease in *d* reflects an animal's tendency to produce smoother paths containing more relatively straight-line movements, where the measured path length decreases slowly with decreasing spatial resolution.

Data were first examined in 30 min time resolutions and analyzed using two-way analysis of variance (ANOVA) with time as a repeated measure. As there were numerous significant interactions with time (data not shown), behaviors were examined in specific time resolutions using two-way ANOVA. Specific post hoc comparisons between selected groups were done using Newman–Keuls. Significance was demonstrated by surpassing an alpha level of 0.05 (two-tailed).

2.4. Procedure

Animals were tested in the dark and during the dark phase of their light cycle. For each experimental session, naive animals were brought to the testing room and allowed to sit for 60 min before being given treatment injections under red lights in the testing room. Test injections were administered 10 min prior to testing in the BPM chamber. Data were collected for 120 min during the dark phase of the animals' light/dark cycle. The chambers were cleaned thoroughly between testing sessions.

2.5. Drugs

Drugs used were: 1(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI; Research Biochemicals, Natick, MA); ketanserin tartrate (Research Biochemicals, Natick, MA) and phencyclidine (PCP; Research Biochemicals, Natick, MA). All drugs were dissolved in saline and administered in a volume of 1 ml/kg subcutaneously. Combinations of drugs were administered in a cocktail preparation in a single injection. Doses are expressed as the salt.

3. Results

3.1. Experiment 1

3.1.1. Procedure

Fifty-two rats were administered either saline or 1.0 mg/kg ketanserin along with either saline, 0.27 mg/kg DOI, or 2.25 mg/kg PCP. The six groups ($n = 8–9$) were tested for 120 min. These doses were based on past experiments (Wing et al., 1990) and pilot studies.

3.1.2. Results

Although these groups were tested together to conserve animals, they were analyzed separately. As we had an a priori hypothesis that ketanserin would antagonize the effects of DOI, but were unsure about the effects of ketanserin on PCP, we analyzed these data sets separately into DOI groups and PCP groups. These data are presented together, in Table 1, to avoid needless repetition of data.

3.1.2.1. DOI. DOI decreased locomotor and investigatory activity and produced smoother locomotor paths. These effects were antagonized by ketanserin pretreatment. There were significant interactions between ketanserin and DOI during the first 30 min of testing for crossings ($F(1, 31) = 11.62$), *d* ($F(1, 31) = 11.87$), holepokes ($F(1, 31) = 21.08$) and rearings ($F(1, 31) = 32.84$). Some of these significant interactions between ketanserin and DOI persisted during the second 30 min of testing, i.e. for cross-

Table 1
Effects of pretreatment with either saline or 1.0 mg/kg ketanserin, combined with treatment of either saline, 0.27 mg/kg DOI, or 2.25 mg/kg PCP, on locomotor and investigatory activity and locomotor patterns

	Crossings		Spatial *d*		Holepokes		Rearings	
	0–30	31–60	0–30	31–60	0–30	31–60	0–30	31–60
Saline	283 ± 11	111 ± 14	1.569 ± 0.013	1.643 ± 0.020	120 ± 10	98 ± 16	119 ± 8	48 ± 8
DOI	164 ± 20[a]	68 ± 9[a]	1.472 ± 0.019[a]	1.636 ± 0.025	28 ± 5[a]	21 ± 8[a]	19 ± 7[a]	11 ± 4[a]
PCP	288 ± 48	411 ± 84[a]	1.644 ± 0.030[a]	1.512 ± 0.040	28 ± 4[a]	39 ± 10[a]	2 ± 2[a]	2 ± 1[a]
Ketanserin	299 ± 36	107 ± 14	1.599 ± 0.009	1.695 ± 0.017	103 ± 9	65 ± 14	121 ± 12	53 ± 11
Ketanserin + DOI	325 ± 13[b]	130 ± 11[b]	1.597 ± 0.022[b]	1.623 ± 0.015	94 ± 11[b]	74 ± 13[b]	123 ± 9[b]	56 ± 10[b]
Ketanserin + PCP	194 ± 38	214 ± 40[b]	1.688 ± 0.018[a]	1.635 ± 0.063	23 ± 5[a]	24 ± 5[a]	0 ± 0[a]	0 ± 0[a]

DOI decreased all behaviors in the initial 30 min and these effects were blocked by ketanserin pretreatment. PCP increased crossings and decreased investigation in the second 30 min. The effect of PCP on locomotor activity was blocked by ketanserin pretreatment. Increases in spatial *d* reflect decreased smoothness of locomotor paths, while decreases in *d* reflect increased smoothness. Data are presented as group means ± S.E.M. for the first and second 30 min of testing.

[a] $P < 0.05$, group is different from saline group.

[b] $P < 0.05$, group is different from its respective treatment alone group.

ings ($F(1, 31) = 7.25$), holepokes ($F(1, 31) = 10.77$) and rearings ($F(1, 31) = 5.45$). Specific comparisons revealed that, in the initial 30 min of testing, DOI treatment significantly decreased crossings, spatial *d*, holepokes and rearings, indicated by [a] in Table 1. Ketanserin, which had no effect by itself, significantly attenuated these effects of DOI, indicated by [b] in Table 1. This pattern of antagonism was also exhibited during the second 30 min of testing for all behaviors, except *d* (Table 1). Thus, 1.0 mg/kg ketanserin was ascertained to be an effective antagonist of the locomotor and investigatory effects of the 5-HT_2 receptor agonist DOI.

3.1.2.2. PCP. PCP increased locomotor activity and decreased investigatory behaviors. Ketanserin reduced the effect of PCP on locomotor activity, without altering the effects of PCP on investigatory behavior. As in the analysis of the DOI data set, these data were examined for the first and second 30 min blocks. There was a significant main effect for PCP during the initial 30 min for *d* ($F(1, 30) = 17.53$), holepokes ($F(1, 30) = 138.96$) and rearings ($F(1, 30) = 291.57$). There were also significant main effects for PCP during the second 30 min of testing for crossings ($F(1, 30) = 16.86$), *d* ($F(1, 30) = 6.04$), holepokes ($F(1, 30) = 16.49$) and rearings ($F(1, 30) = 56.44$). Specific comparisons revealed that PCP treatment significantly increased crossings and decreased holepokes and rearings during the second 30 min (see Table 1). Ketanserin reduced the effect of PCP on crossings, as suggested by a trend for an interaction between PCP and ketanserin for crossings ($F(1, 30) = 3.77$; $p = 0.06$). Based on this trend and the a priori hypothesis, post-hoc comparisons were used to confirm that the ketanserin + PCP group differed significantly from the PCP alone group for crossings in the second 30 min of testing (Table 1). In contrast, the same dose of ketanserin that robustly antagonized all the effects of the 5-HT_2 receptor agonist DOI, failed to significantly attenuate the effects of PCP on spatial d and investigatory activity.

3.2. Experiment 2

3.2.1. Procedure

One hundred and thirty-eight rats were treated with either saline, 0.75, 2.25, 6.75, or 10.125 mg/kg PCP, 0.27 mg/kg DOI, or combinations thereof. The ten groups ($n = 8–25$) were tested for 120 min. Data from seven rats were lost due to technical problems. Data from the saline, DOI and PCP 2.25 groups were shared, in part, with Experiment 1. These drug doses were selected on the basis of Experiment 1 and past experiments (Wing et al., 1990; Lehmann-Masten and Geyer, 1991).

3.2.2. Results

As in the preceding experiments, DOI decreased locomotor and investigatory behaviors while PCP increased locomotor and decreased investigatory responses while producing smoother locomotor paths. Complex interactions between these effects were observed, especially for measures of locomotor activity. As in the above experiments, the first 30 min block of testing was determined to be the most sensitive for the effect of DOI, while the second 30 min block was determined to be the most sensitive time resolution for the effect of PCP. Accordingly, these time resolutions were examined further. There were significant interactions between DOI and PCP in the first 30 min of testing for crossings ($F(4, 121) = 14.31$), *d* ($F(4, 121) = 12.82$), holepokes ($F(4, 121) = 14.07$) and rearings ($F(4, 121) = 17.30$). There were also significant interactions between DOI and PCP in the second 30 min of testing for crossings ($F(4, 121) = 18.31$), spatial *d* ($F(4, 121) = 13.15$) and holepokes ($F(4, 121) = 7.01$).

Specific comparisons revealed that the acute administration of 2.25 and 6.75 mg/kg PCP significantly increased crossings in the second 30 min of testing, indicated by the asterisk (*) symbol (Fig. 1). No dose of PCP affected crossings during the initial 30 min. The effect of PCP on spatial *d* was somewhat more complex. In the first 30 min, 6.75 and 10.125 mg/kg PCP increased *d* and, thus, increased the roughness of the locomotor path. For the 10.125 mg/kg dose of PCP, this effect persisted into the second 30 min. These changes in locomotor patterns can be noted in plots of rats representative of the 6.75 and 10.125 mg/kg PCP groups (Fig. 3C and E). Conversely, the 2.25 mg/kg dose of PCP decreased spatial *d* during the second 30 min of testing. While these same data were not significant in the context of experiment 1 (Table 1), this biphasic effect of PCP has been noted before (Masten, personal communication). PCP also decreased holepokes

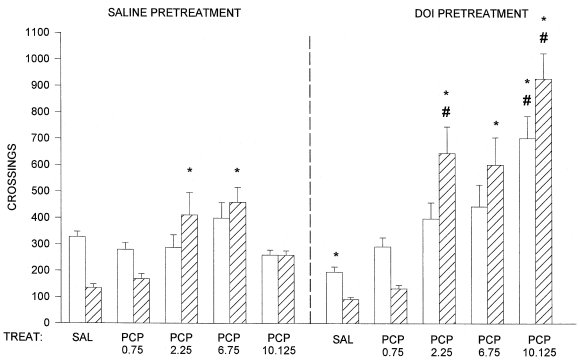

Fig. 1. 2.25 and 6.75 mg/kg PCP treatment significantly increased crossings, a measure of locomotor activity, during the second 30 min of testing. This hyperactivity was not evident in the 10.125 mg/kg PCP group, probably due to the ataxia caused by this high dose of PCP. 0.27 mg/kg DOI pretreatment produced hyperactivity in the 2.25–10.125 mg/kg PCP groups. Data are presented as group means ± S.E.M. for the first 30 min (open bars) and the second 30 min (hatched bars) of testing. * $P < 0.05$, group is different from saline group. # $P < 0.05$, group is different from its respective PCP treatment alone group.

Table 2
Effect of pretreatment with either saline or 0.27 mg/kg DOI, combined with treatment of either saline or 0.75–10.125 mg/kg PCP, on investigatory activity

	Holepokes		Rearings	
	0–30	31–60	0–30	31–60
Saline	128±10	109±10	88±9	26±5
PCP (0.75 mg/kg)	69±12[a]	92±17	17±4[a]	18±4
PCP (2.25 mg/kg)	28±4[a]	39±10[a]	2±2[a]	2±1[a]
PCP (6.75 mg/kg)	37±10[a]	32±5[a]	1±1[a]	5±3[a]
PCP (10.125 mg/kg)	71±11[a]	56±6[a]	0±0[a]	10±4
DOI	35±4[a]	32±5[a]	22±5[a]	12±3
DOI+PCP (0.75 mg/kg)	68±11[a]	56±18[a]	34±11[a]	12±6
DOI+PCP (2.25 mg/kg)	36±8[a]	37±14[a]	34±1[a]	1±1[a]
DOI+PCP (6.75 mg/kg)	30±4[a]	34±8[a]	1±0[a]	3±2[a]
DOI+PCP (10.125 mg/kg)	30±5[a]	46±6[a]	12±5[a]	11±5

Data are presented as group means±S.E.M. for the first and second 30 min of testing.
[a] $P < 0.05$, group is different from saline group.

and rearings (see Table 2). Thus, PCP increased locomotor activity. Independent of the effect on locomotor activity, PCP produced biphasic effects on locomotor patterns. 2.25 mg/kg PCP produced smoother locomotor paths, while higher doses decreased the smoothness of the locomotor path. PCP also decreased investigatory activity.

DOI pretreatment significantly decreased crossings, rearings and spatial *d*, but only during the first 30 min (Figs. 1 and 2, Table 2). DOI decreased holepokes over the entire hour (Table 2). DOI pretreatment, which had no effect on locomotion on its own during the second 30 min, appeared to further increase the effect of some doses of PCP on crossings and spatial *d*. The PCP2.25 group, which showed increased crossings (Fig. 1), showed more activity when DOI pretreatment was added, reflected in a significant difference between the PCP2.25 group and the DOI + PCP2.25 group (indicated by the pound [#] symbol in Fig. 1). The PCP6.75 group was also significantly different from saline, but interpreting any interaction of this dose of PCP with DOI pretreatment is hampered by the high level of activity produced by this dose of PCP. Thus, while the PCP6.75 group was not significantly different from the DOI + PCP6.75 group, this result was probably due to the already high level of activity produced by 6.75 mg/kg PCP. Interestingly, DOI produced a robust increase in crossings even when PCP did not produce hyperactivity, but only at a high dose of PCP. That is, the PCP10.125 group was not significantly different from saline. This high PCP dose appears to be towards the end of the dose response curve, in which ataxia and diminished activity occur. When DOI pretreatment was added to the 10.125 mg/kg dose of PCP, crossings were increased significantly, as indicated by the pound symbol in Fig. 1. DOI pretreatment had no effect, however, on the lowest dose of 0.75 mg/kg PCP, a dose which was also not significantly different from saline.

The acute administration of 2.25 mg/kg PCP significantly decreased spatial *d* during the second 30 min (Fig. 2). DOI pretreatment, which again had no effect on its own in this time resolution, appeared to further decrease spatial *d* in rats treated with 2.25 mg/kg PCP. Higher doses of PCP, however, increased *d* and produced rougher locomotor paths. DOI pretreatment decreased *d* in these groups as well, indicated by the significant difference between the PCP6.75 and DOI + PCP6.75 groups and between the PCP10.125 and DOI + PCP10.125 groups (indicated by the pound symbol in Fig. 2). The effect of DOI on PCP-induced changes in locomotor patterns can be noted in the plots of rats representative of these groups (Fig. 3D and F). DOI pretreatment again had no effect on the 0.75 mg/kg dose of PCP, a dose which was not significantly different from saline.

Specific comparisons revealed that PCP decreased holepokes and rearings in both blocks of testing (Table 2). There was no significant difference, however, between any of the PCP alone groups and their respective DOI + PCP groups, which indicates that DOI pretreatment did not influence the effect of PCP on investigatory activity as it did on locomotor activity. Thus, despite the significant interactions between DOI and PCP for investigatory activity, specific comparisons revealed that PCP did not produce further attenuations in investigation in DOI-pretreated animals and, thus, DOI did not potentiate the effects of PCP. The possibility that DOI may have reduced the effects of PCP cannot be assessed because DOI alone suppressed investigatory activity.

Thus, the interaction between DOI and multiple doses of PCP suggests that the combination of these two drugs produces a behavioral profile characterized by profoundly

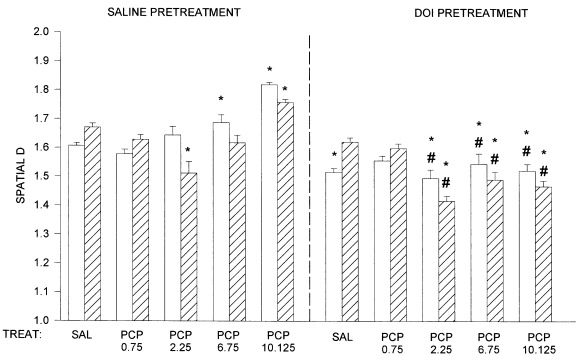

Fig. 2. PCP produced biphasic effects on locomotor patterns. 6.75 and 10.125 mg/kg PCP treatment significantly increased *d*, while 2.25 mg/kg PCP decreased *d* during the second 30 min. 0.27 mg/kg DOI pretreatment produced decreases in *d* for 2.25–10.125 mg/kg PCP groups. Increases in spatial *d* reflect increased roughness of locomotor paths, while decreases in *d* reflect increased smoothness. Data are presented as group means±S.E.M. for the first 30 min (open bars) and the second 30 min (hatched bars) of testing. * $P < 0.05$, group is different from saline group. # $P < 0.05$, group is different from its respective PCP treatment alone group.

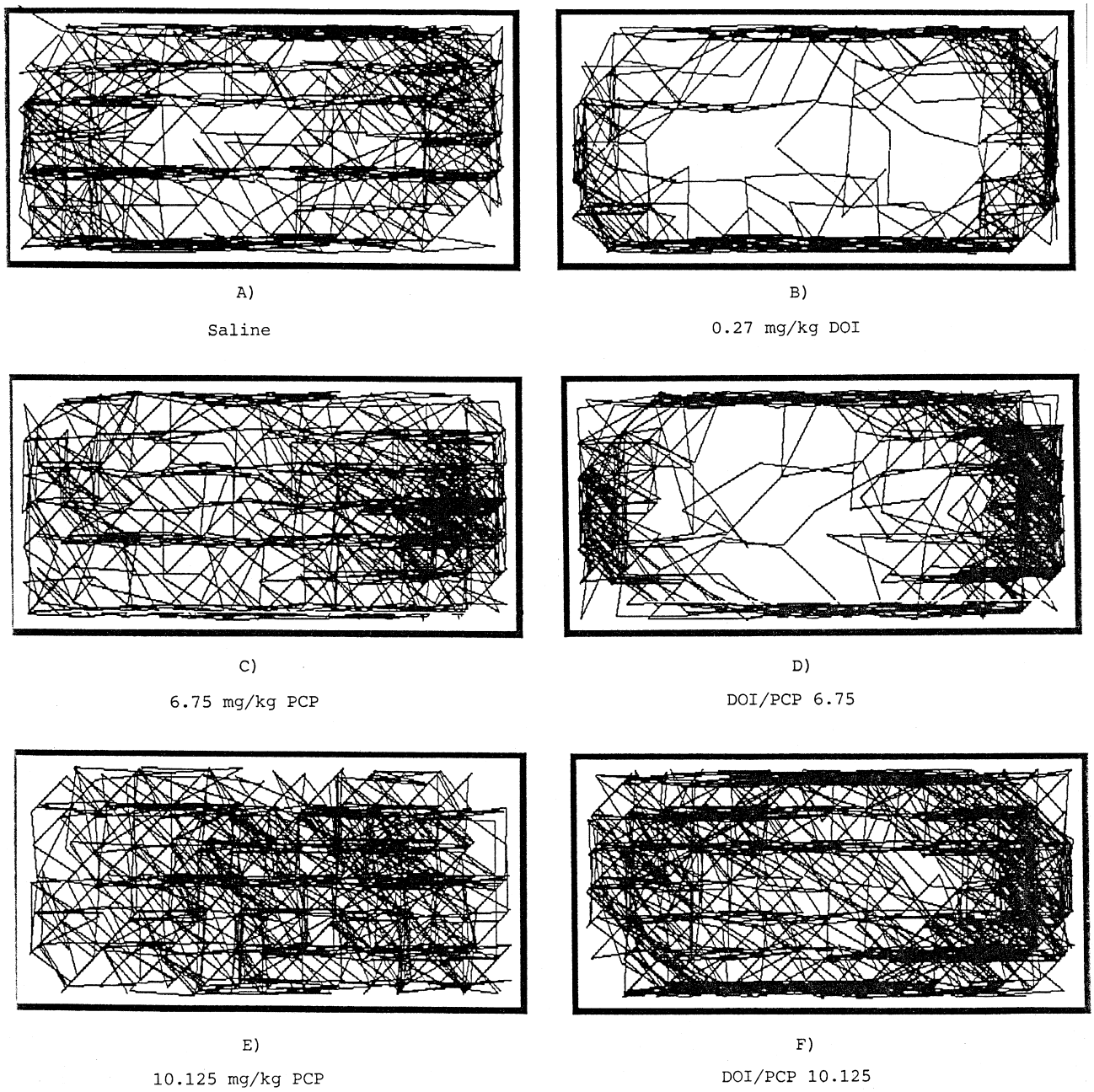

Fig. 3. Effects of DOI and PCP on the spatial patterning of exploration during the initial 30 min of testing. This time resolution was chosen for its ability to represent changes in locomotor pattern while minimizing representation of confounding factors of locomotor activity increases. Shown are computer-generated plots of $X–Y$ position changes of individual, representative rats administered: (A) saline, (B) 0.27 mg/kg DOI, (C) 6.75 mg/kg PCP, (D) DOI + PCP6.75, (E) 10.125 mg/kg PCP and (F) DOI + PCP10.125. In contrast to the varied path with which the saline-treated rat (A) explored the entire chamber, the DOI-treated rat (B) explored the periphery of the chamber and made relatively fewer entries into the center. Rats treated with PCP (C and E) demonstrated more varied behavior with rougher paths, compared to the saline-treated animal. DOI pretreatment altered these PCP-induced patterns of activity by producing smoother paths (D and F).

enhanced locomotor activity and altered locomotor patterns.

4. Discussion

The results of these studies provide converging evidence of complex functional interactions between serotonergic and glutamatergic systems in the modulation of spontaneous locomotor behavior in rats. The robust potentiation of the locomotor-activating effects of PCP by the 5-HT_2 receptor agonist DOI, in the absence of any potentiation of the effects of PCP on investigatory behaviors and in a manner that is inconsistent with a simple shift in the dose–effect functions for either PCP or DOI, is a new

finding that extends previous studies of the influences of 5-HT_2 receptor antagonists on PCP effects. Collectively, these observations support the hypothesis that serotonergic manipulations alter the behavioral effects of PCP by virtue of interactions mediated through circuit connections rather than more direct intra-synaptic interactions. Corroborating previous reports, the 5-HT_2 receptor antagonist ketanserin was able to block the full range of behavioral effects of the 5-HT_2 receptor agonist DOI completely and to reduce only some of the effects of PCP. As ketanserin is somewhat selective for 5-HT_{2A} over 5-HT_{2C} receptors (Hoyer and Schoeffter, 1991), this interaction may be due to preferential antagonism of 5-HT_{2A} receptors; but the dose of ketanserin used was high enough to make selectivity doubtful. Hence, this interaction is best characterized as related to nonselective 5-HT_2 antagonism. The interaction between the 5-$HT_{2A/2C}$ agonist DOI (Glennon et al., 1992) and PCP, however, was not interpretable as either a simple additivity of the separate effects of the two drugs or a shift in the dose–effect function for PCP. Instead, examination of the interaction between DOI and multiple doses of PCP on the behavioral profile provided by the Behavioral Pattern Monitor strongly suggests that the combination of these two drugs produces a behavioral profile that is not predicted by the effects of either drug by itself.

Acute PCP administration produced an increase in locomotor activity that began to fall off at the highest dose of PCP. PCP had biphasic effects on locomotor patterns. Higher doses of PCP decreased the smoothness of the locomotor path, while a lower dose of 2.25 mg/kg PCP increased the smoothness of the path. PCP also decreased investigatory activity. Some of these effects were altered profoundly by the concomitant activation of 5-HT_2 receptors produced by DOI pretreatment.

It does not appear that this interaction reflects a shift of the PCP dose–response curve, for several reasons. First, the highest dose of PCP produced ataxia and, thus, a decrease in locomotor activity and rougher locomotor paths (Figs. 1 and 2). A shift of the dose–response curve to the right would be expected to decrease activity and increase the roughness of the locomotor path. DOI pretreatment, however, produced even higher levels of activity and even smoother paths in PCP-treated rats. Second, there was no interaction between DOI and the 0.75 mg/kg dose of PCP for any measure. If DOI pretreatment were shifting the dose–response curve of PCP to the right, it would be expected that the addition of DOI to the behaviorally ineffective 0.75 mg/kg dose of PCP would increase activity. The fact that DOI had no effect on 0.75 mg/kg PCP supports the hypothesis that DOI did not shift the PCP curve to the right. Third, if DOI pretreatment were shifting the dose–response curve of PCP to the left, it would be expected that the addition of DOI to the 2.25 mg/kg dose of PCP would produce a behavioral profile similar to that of rats treated with 0.75 mg/kg PCP. The fact that the DOI + PCP2.25 group exhibited increases in activity, very much unlike the PCP0.75 group, supports the hypothesis that DOI did not shift the PCP curve to the left.

DOI did not potentiate the effects of PCP treatment on investigatory behavior. DOI pretreatment, which decreased holepokes and rearings by itself, did not alter the PCP-induced suppression of investigatory activity. This finding is consistent with the observation that the 5-HT_2 receptor antagonist ketanserin also failed to alter the effects of PCP on investigatory behavior, despite decreasing the effect of PCP on locomotor activity. Because of the significant suppression of investigatory activity produced by DOI alone, however, it would be difficult to observe if DOI reduced the effects of PCP. Nevertheless, DOI did not produce any further decreases in investigation in PCP-treated rats. The lack of a robust effect of DOI pretreatment on PCP-treated rats for investigation suggests that the interaction between DOI and PCP is evident for locomotor activity and patterns, but relatively absent for investigatory activity.

Based on our knowledge of the effects of DOI on locomotor and investigatory activity, reported elsewhere (Wing et al., 1990), the combined effects of DOI and PCP do not reflect a simple shift in the DOI dose–response function. Throughout a wide dose range, DOI produces a linear suppression of locomotor activity and investigatory holepokes and rearings (Wing et al., 1990). Thus, the large increases in locomotor activity observed with combinations of DOI and PCP are inconsistent with a shift in the DOI curve to either the right or the left. Similarly, administration of PCP did not alter the suppression of investigatory activity produced by DOI, which does not support a simple shift in the DOI dose–effect curve.

Thus, DOI administration interacted with PCP treatment to produce a new effect characterized by a specific behavioral profile. Whatever the mechanism for DOI modulation of the effects of PCP on locomotor activity, it appears that 5-HT_2 receptor agonism enhances the PCP-induced increases in locomotor activity if, and only if, there is a sufficient concentration of PCP extant. The overriding effect of DOI pretreatment to increase locomotion, despite the behavioral effect of PCP alone, was mirrored in the spatial *d* measurement. As these effects, in toto, did not reflect a shift in the PCP dose–response curve, it appears that the co-administration of DOI and PCP resulted in a new profile of behavior, characterized by changes in the amount and pattern of locomotor activity.

The specific mechanism mediating the interaction of PCP and DOI is not known. The facts are inconsistent with an interaction at a particular receptor or class of synapses. Rather, it seems more likely that these interactions reflect changes in the dynamics of neuronal circuits in which both glutamatergic and serotonergic systems are involved. One possible example of such a circuit interaction might be one of the putative circuits between the limbic system and the motor system, which are routed through the nucleus accumbens and globus pallidus (for review see Mogenson et

al., 1980). For example, there are glutamatergic projections from the hippocampus to the nucleus accumbens and both of these areas are known to contain 5-HT_2 receptors (Fonnum et al., 1979; Mogenson et al., 1980; Mogenson and Nielsen, 1984; Palacios and Dietl, 1988; Morilak et al., 1993). It is conceivable that agonism of either of these receptor populations by DOI might interact with the glutamatergic influence of PCP in this circuit. This hypothesis is speculation at this point and is only one of many possible explanations of how DOI and PCP interact to produce a new profile of locomotor behavior.

In conclusion, the effects of PCP on locomotor activity were altered by 5-HT_2 receptor antagonism. 5-HT_2 receptor agonism altered not only the effects of PCP on locomotor activity, but on locomotor patterns as well. Neither 5-HT_2 receptor antagonism or agonism, however, modified the effects of PCP on investigatory behavior. These results indicate that locomotor activity, rather than investigatory behavior, is more sensitive to the proposed circuit interactions between serotonin and glutamate.

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References

Ali, S.F., Newport, G.D., Bracha, H.S., 1994. Phencyclidine and (+)-MK-801-induced circling preference: Correlation with monoamine levels in striatum of the rat brain. Neurotoxicol. Teratol. 16, 335–342.

Carlsson, M.L., 1995. The selective 5-HT_{2A} receptor antagonist MDL 100,907 counteracts the psychomotor stimulation ensuing manipulations with monoaminergic, glutamatergic or muscarinic neurotransmission in the mouse-implications for psychosis. J. Neural Transm. 100, 225–237.

Castellani, S., Adams, P.M., 1981. Effects of dopaminergic drugs on phencyclidine-induced behavior in the rat. Neuropharmacology 20, 371–374.

Corbett, R., Camacho, F., Woods, A.T., Kerman, L.L., Fishkin, R.J., Brooks, K., Dunn, R.W., 1995. Antipsychotic agents antagonize non-competitive *N*-methyl-D-aspartate antagonist-induced behaviors. Psychopharmacology 120, 67–74.

Domino, E.F., Luby, E.D., 1981. Abnormal mental states induced by phencyclidine as a model of schizophrenia. In: Domino, E.F. (Ed.), PCP (Phencyclidine): Historical and Current Perspectives. NPP Books, Ann Arbor, MI, pp. 400–418.

Fonnum, F., Karlsen, R.L., Mathe-Sorenssen, P., Skrede, K.K., 1979. Localization of neurotransmitters, particularly glutamate in hippocampus, septum, nucleus accumbens and superior colliculus. Prog. Brain Res. 51, 169–191.

Garey, R.E., 1979. PCP (phencyclidine): An update. J. Psychedelic Drugs 11, 265–275.

Garey, R.E., Heath, R.G., 1976. The effects of phencyclidine on the uptake of ^{3}H-catecholamines by rat striatal and hypothalamic synaptosomes. Life Sci. 18, 1105–1110.

Geyer, M.A., 1990. Approaches to the characterization of drug effects on locomotor activity in rodents. In: Adler, M.W., Cowan, A. (Eds.), Modern Methods in Pharmacology: Testing and Evaluation of Drugs of Abuse. Wiley-Liss, New York, pp. 81–99.

Geyer, M.A., Russo, P.V., Masten, V.L., 1986. Multivariate assessment of locomotor behavior: Pharmacological and behavioral analyses. Pharmacol. Biochem. Behav. 25, 277–288.

Gleason, S.D., Shannon, H.E., 1997. Blockade of phencyclidine-induced hyperlocomotion by olanzepine, clozapine and serotonin receptor subtype selective antagonists in mice. Psychopharmacology 129, 79–84.

Glennon, R.A., Raghupathi, R., Bartyzel, P., Teitler, M., Leonhardt, S., 1992. Binding of phenalkylamine derivatives at 5-HT1C and 5-HT2 serotonin receptors: Evidence for a lack of selectivity. J. Med. Chem. 35, 734–740.

Hernandez, L., Auerbach, S., Hoebel, B.G., 1988. Phencyclidine (PCP) injected in the nucleus accumbens increases extracellular dopamine and serotonin as measured by microdialysis. Life Sci. 42, 1713–1723.

Hertel, P., Mathé, J.M., Nomikos, G.G., Iurlo, M., Mathé, A.A., Svensson, T.H., 1996. Effects of D-amphetamine and phencyclidine on behavior and extracellular concentrations of neurotensin and dopamine in the ventral striatum and the medial prefrontal cortex of the rat. Behav. Brain Res. 72, 103–114.

Hori, T., Suzuki, T., Baba, A., Abe, S., Yamamoto, T., Moroji, T., Shiraishi, H., 1996. Effects of phencyclidine metabolites on serotonin uptake in rat brain. Neurosci. Lett. 209, 153–156.

Hoyer, D., Schoeffter, P., 1991. 5-HT receptors: Subtypes and second messengers. J. Receptor Res. 11, 197–214.

Jackson, D.M., Johansson, C., Lindgren, L.-M., Bengtsson, A., 1994. Dopamine receptor antagonists block amphetamine and phencyclidine-induced motor stimulation in rats. Pharmacol. Biochem. Behav. 48, 465–471.

Javitt, D.C., Zukin, S.R., 1991a. Recent advances in the phencyclidine model of schizophrenia. Am. J. Psychiatry 10, 1301–1308.

Javitt, D.C., Zukin, S.R., 1991b. Mechanisms of phencyclidine (PCP)-*N*-methyl-D-aspartate (NMDA) receptor interaction: Implications for schizophrenia. In: Tamminga, C.A., Schulz, S.C. (Eds.), Advances in Neuropsychiatry and Psychopharmacology, vol. 1: Schizophrenia Research. Raven Press, New York, NY, pp. 13–19.

Johnson, K.M., Jones, S.M., 1990. Neuropharmacology of phencyclidine: Basic mechanisms and therapeutic potential. Annu. Rev. Pharmacol. Toxicol. 30, 707–750.

Kameyama, T., Sugimoto, A., Yamaguchi, K., Hiramatsu, M., Furukawa, H., Nabeshima, T., 1988. The relationship between phencyclidine-induced stereotyped behavior and dopamine release using in vivo voltammetry. Res. Comm. Substance Abuse 9, 89–105.

Kitaichi, K., Yamada, K., Hasegawa, T., Furukawa, H., Nabeshima, T., 1994. Effects of risperidone on phencyclidine-induced behaviors: Comparison with haloperidol and ritanserin. Jpn. J. Pharmacol. 66, 181–189.

Lapin, I.P., Rogawski, M.A., 1995. Effects of D1 and D2 dopamine receptor antagonists and catecholamine depleting agents on the locomotor stimulation induced by dizocilpine in mice. Behav. Brain Res. 70, 145–151.

Lehmann-Masten, V.D., Geyer, M.A., 1991. Spatial and temporal patterning distinguishes the locomotor activating effects of dizocilpine and phencyclidine in rats. Neuropharmacology 30, 629–636.

Lillrank, S.M., O'Connor, W.T., Oja, S.S., Ungerstedt, U., 1994. Systemic phencyclidine administration is associated with increased dopamine, GABA, and 5-HIAA levels in the dorsolateral striatum of conscious rats: An in vivo microdialysis study. J. Neural Transm. 95, 145–155.

Luby, E.D., Cohen, B.D., Rosenbaum, G., Gottlieb, J.S., Kelly, R., 1959. Study of a new schizophrenomimetic drug: Sernyl. A.M.A. Arch. Neurol. Psychiatry 81, 363–369.

Luby, E.D., Gottlieb, J.S., Cohen, B.D., Rosenbaum, G., Domino, E.F., 1962. Model psychoses and schizophrenia. Am. J. Psychiatry 119, 61–65.

Maurel-Remy, S., Bervoets, K., Millan, M.J., 1995. Blockade of phencyclidine-induced hyperlocomotion by clozapine and MDL 100,907 in rats reflects antagonism of 5-HT_{2A} receptors. Eur. J. Pharmacol. 280, R9–R11.

Mogenson, G.J., Nielsen, M., 1984. A study of the contribution of hippocampal–accumbens–subpallidal projections to locomotor activity. Behav. Neur. Biol. 42, 38–51.

Mogenson, G.J., Jones, D.L., Yim, C.Y., 1980. From motivation to action: Functional interface between the limbic system and the motor system. Prog. Neurobiol. 14, 69–97.

Morilak, D.A., Garlow, S.J., Ciaranello, R.D., 1993. Immunocytochemical localization and description of neurons expressing serotonin$_2$ receptors in the rat brain. Neuroscience 54, 701–717.

Murray, T.F., Horita, A., 1979. Phencyclidine-induced stereotyped behavior in rats: Dose–response effects and antagonism by neuroleptics. Life Sci. 24, 2217–2226.

Nabeshima, T., Ishikawa, K., Yamaguchi, K., Furukawa, H., Kameyama, T., 1987a. Phencyclidine-induced head-twitch responses as 5-HT_2 receptor-mediated behavior in rats. Neurosci. Lett. 76, 335–338.

Nabeshima, T., Yamaguchi, K., Ishikawa, K., Furukawa, H., Kameyama, T., 1987b. Potentiation in phencyclidine-induced serotonin-mediated behaviors after intracerebroventricular administration of 5,7-dihydroxytryptamine in rats. J. Pharmacol. Exp. Ther. 243, 1139–1146.

Ögren, S.O., Goldstein, M., 1994. Phencyclidine- and dizocilpine-induced hyperlocomotion are differently mediated. Neuropsychopharmacology 11, 167–177.

Palacios, J.M., Dietl, M.M., 1988. Autoradiographic studies of serotonin receptors. In: Sanders-Bush, E. (Ed.), The Serotonin Receptor. Humana Press, New York, pp. 89–138.

Paulus, M.P., Geyer, M.A., 1991. A temporal and spatial scaling hypothesis for the behavioral effects of psychostimulants. Psychopharmacology 104, 6–16.

Rao, T.S., Kim, H.S., Lehmann, J., Martin, L.L., Wood, P.L., 1989. Differential effects of phencyclidine (PCP) and ketamine on mesocortical and mesostriatal dopamine release in vivo. Life Sci. 45, 1065–1072.

Smith, R.C., Meltzer, H.Y., Arora, R.C., Davis, J.M., 1977. Effects of phencyclidine on [^{3}H]catecholamine and [^{3}H]serotonin uptake in synaptosomal preparations from rat brain. Biochem. Pharmacol. 26, 1435–1439.

Snyder, S.H., 1988. Psychotogenic drugs as models for schizophrenia. Neuropsychopharmacology 1, 197–199.

Steinpreis, R.E., Salamone, J.D., 1993. The role of nucleus accumbens dopamine in the neurochemical and behavioral effects of phencyclidine: A microdialysis and behavioral study. Brain Res. 612, 263–270.

Svensson, T.H., Mathé, J.M., Andersson, J.L., Nomikos, G.G., Hildebrand, B.E., Marcus, M., 1995. Mode of action of atypical neuroleptics in relation to the phencyclidine model of schizophrenia: Role of 5-HT_2 receptor and α_1-adrenoceptor antagonism. J. Clin. Psychopharmacol. 15, 11S–18S.

Tsutsumi, T., Hirano, M., Matsumoto, T., Nakamura, K., Hashimoto, K., Hondo, H., Yonezawa, Y., Tsukashima, A., Nakane, H., Uchimura, H., Nakahara, T., 1995. Involvement of dopamine D_1 receptors in phencyclidine-induced behavioral stimulation in rats. Clin. Neuropharmacol. 18, 64–71.

Wing, L.L., Tapson, G.S., Geyer, M.A., 1990. 5-HT2 mediation of acute behavioral effects of hallucinogens in rats. Psychopharmacology 100, 417–425.

Yamaguchi, K., Nabeshima, T., Kameyama, T., 1986. Role of dopaminergic and serotonergic neuronal systems in the prefrontal cortex of rats in phencyclidine-induced behaviors. J. Pharmacobio-Dyn. 9, 987–996.

Yamaguchi, K., Nabeshima, T., Ishikawa, K., Yoshida, S., Kameyama, T., 1987. Phencyclidine-induced head-weaving and head-twitch through interaction with 5-HT_1 and 5-HT_2 receptors in reserpinized rats. Neuropharmacology 26, 1489–1497.