0022-3565/88/2463-0924$02.00/0
THE JOURNAL OF PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS
Vol. 246, No. 3

Printed in U.S.A.

Lysergic Acid Diethylamide and 2,5-Dimethoxy-4-methylamphetamine are Partial Agonists at Serotonin Receptors Linked to Phosphoinositide Hydrolysis

ELAINE SANDERS-BUSH, KEVIN D. BURRIS and KAREN KNOTH
Departments of Pharmacology and Psychiatry, Vanderbilt University School of Medicine, Nashville, Tennessee
Accepted for publication May 26, 1988

ABSTRACT

Based on electrophysiological, radioligand binding, and behavioral studies in laboratory animals, it is generally believed that the psychotomimetic effects of the phenethylamine and indolealkylamine hallucinogens are mediated by central serotonin (5-HT) receptors, in particular the 5-HT-2 subtype. Agonist-stimulated phosphoinositide hydrolysis was utilized to determine the potency and efficacy of racemic 1-(2,5)-dimethoxy-4-methylphenyl)-2-aminopropane (DOM), and *d*-lysergic acid diethylamide (LSD) at the 5-HT-2 receptor in rat cerebral cortex and the 5-HT-1c receptor in rat choroid plexus. Both DOM and LSD stimulated phosphoinositide hydrolysis in cerebral cortex. These effects were blocked by the 5-HT-2 antagonists, ketanserin and spiperone, but not by antagonists of muscarinic, *alpha*-1 adrenergic or histaminergic receptors. The maximum responses of DOM and LSD, respectively, were 76% and 25% of the maximum response to 5-HT. However, LSD was 500 times more potent than was racemic DOM. Consistent with a partial agonist effect, LSD partially blocked the effect of 5-HT, with a maximal inhibition equivalent to the intrinsic activity of LSD alone. In choroid plexus, DOM and LSD stimulated phosphoinositide hydrolysis and both responses were blocked by mianserin and less effectively by spiperone. The maximum effect of DOM was 67% of that of 5-HT, whereas the maximum effect of LSD was only 34% of the maximum response of 5-HT. LSD was 50 times more potent than was racemic DOM. LSD partially antagonized the effect of 5-HT in the choroid plexus, consistent with a partial agonist effect at the 5-HT-1c receptor in this tissue. The isomers of DOM were also evaluated; the R(−) isomer was more potent both in cortex and in choroid plexus. In conclusion, this study demonstrates a direct agonist action of DOM and LSD at 5-HT-2 receptors in the cerebral cortex and 5-HT-1c receptors in the choroid plexus and shows that, in both systems, the hallucinogens function as partial agonists.

LSD, an indolealkylamine, and DOM, a phenethylamine, are representatives of the two major classes of hallucinogenic drugs. Electrophysiological, behavioral and radioligand binding studies in laboratory animals suggest that hallucinogens interact potently with central 5-HT receptors. Systemic administration of LSD or DOM to rats decreases the spontaneous activity of LC neurons and increases activation of LC neurons by sciatic nerve stimulation (Rasmussen and Aghajanian, 1986). These effects are reversed by the 5-HT-2 antagonist LY53857 and the potencies of the hallucinogens correlate with their affinities at 5-HT-2 binding sites (Rasmussen *et al.*, 1986). DOM excites facial motoneurons, and this effect is apparently mediated by activation of the 5-HT-2 receptors (Penington and Reiffenstein, 1986). Behavioral studies in rats have demonstrated that LSD and DOM disrupt bar press responding, an effect blocked by the 5-HT-2 antagonist pirenperone (Mokler *et al.*, 1985). Additional behavioral studies showed that the 5-HT-2 antagonist ritanserin blocks LSD/saline discrimination (Colpaert *et al.*, 1985), suggesting that the discriminative cue properties of LSD are related to 5-HT-2 receptor activation. In recent radioligand binding studies, an excellent correlation was found between the affinities of hallucinogens at the 5-HT-2 binding site and their potencies to substitute for DOM as a discriminative cue (Glennon *et al.*, 1984). Furthermore, the affinities at the 5-HT-2 binding site correlated with potencies as hallucinogens in humans (Glennon *et al.*, 1984). This evidence has led to the hypothesis that central 5-HT-2 receptors are involved in the mechanism of the hallucinogenic action of these drugs. Although this speculation may indeed prove to be true, the direct effects of hallucinogenic drugs at central 5-HT-2 receptors are yet to be established.

Electrophysiological and behavioral effects reflect changes distal to the site of interaction of the drug with its receptor; therefore, determination of a drug's actual efficacy or potency at a specific receptor is not possible with these experimental

Received for publication March 4, 1988.

ABBREVIATIONS: LSD, *d*-lysergic acid diethylamide; DOM, 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane; 5-HT, serotonin; LC, locus coeruleus; IP, inositol-monophosphate.

designs. Radioligand binding studies give a measure of the affinity of a drug for specific receptor subtypes, but do not yield information about the mechanism of the drug's action at the receptor. The conclusion that hallucinogenic effects are mediated by central 5-HT-2 receptors is complicated further by the fact that LSD competes with high affinity for five major 5-HT receptor subtypes: 5-HT-1a, 5-HT-1b, 5-HT-1c, 5-HT-1d and 5-HT-2 (Engel *et al.*, 1986; Peroutka, 1986). Functional studies of these 5-HT receptor subtypes are necessary to determine the efficacy and potency of LSD and other hallucinogens at each site and to categorize the drugs as agonists, partial agonists or antagonists. Phosphoinositide hydrolysis is one of the major effector systems linked to central 5-HT receptors (Sanders-Bush and Conn, 1987). Studies have demonstrated clearly that the 5-HT-2 receptor in rat cerebral cortex (Conn and Sanders-Bush, 1984, 1985, 1986a) and the 5-HT-1c receptor in choroid plexus (Conn *et al.*, 1986; Conn and Sanders-Bush, 1986b) are coupled positively to phosphoinositide hydrolysis. Using this functional measure, the direct effects of the indolealkylamine hallucinogen, LSD, and the phenethylamine hallucinogen, DOM, were examined at central 5-HT-2 and 5-HT-1c receptors in cortex and choroid plexus, respectively.

Methods

Drugs. Mianserin HCl and spiperone (free base) were obtained from Research Biochemicals, Inc. (Nattick, MA); 5-HT creatinine sulfate was from Sigma Chemical Co. (St. Louis, MO). The following drugs were kindly donated by the indicated manufacturers: ketanserin tartrate by Janssen Pharmaceutica (Beerse, Belgium) and pargyline HCl by Abbott Laboratories (North Chicago, IL). [^{3}H]myoinositol (15 Ci/mmol) was purchased from American Radiolabeled Chemicals (St. Louis, MO) and stored at 5°C with a small amount of Dowex-1 anion exchange resin in the formate form in order to maintain purity. *d*-LSD tartrate, racemic DOM HCl, *S*(+)-DOM HCl and *R*(−)-DOM HCl were obtained from the National Institute of Drug Abuse (Rockville, MD).

Phosphoinositide hydrolysis. Male Sprague-Dawley rats, 180 to 200 g, were obtained from Sasco, Inc. (Omaha, NE). Agonist-induced phosphoinositide hydrolysis in cerebral cortex slices was determined by a modification of the method of Berridge *et al.*, (1982) as described previously (Conn and Sanders-Bush, 1985). In this method, the formation of [^{3}H]IP in the presence of lithium is used as an index of phosphoinositide hydrolysis. Briefly, cerebral cortex was sliced, the slices washed gently and incubated in Krebs-bicarbonate buffer for 30 min in an atmosphere of 95% O_2-5% CO_2. The slices were then labeled by incubating with [^{3}H]inositol (1 μCi/ml) for 2 hr. LiCl (10 mM) and pargyline (10 μM) were added and 15 min later, the assay was initiated by the addition of agonist. After a 45-min incubation with agonist, the reaction was terminated by the addition of chloroform/methanol. After extraction, an aliquot of the aqueous phase containing the inositol phosphates was transferred to a Dowex-1 anion exchange resin column. Inositol was eluted and discarded and [^{3}H]IP was eluted with ammonium formate/formic acid and counted.

Choroid plexi were obtained from 20-day old male Sprague-Dawley rats (Sasco, Inc.). After hemisecting the brain, the choroid plexi in the lateral ventricles were exposed by gently pushing aside the hippocampus. With gentle pulling, the plexi in the lateral ventricle and third ventricle were removed as one. The phosphoinositide hydrolysis assay was the same as in cortex, except that each tube contained one whole, unsliced choroid plexus (weighing approximately 0.5 mg) and the incubation duration for [^{3}H]inositol labeling was 90 min.

Results

Effects of LSD and DOM in cerebral cortex. Racemic DOM increased phosphoinositide hydrolysis in slices of cerebral cortex in a concentration-dependent manner (fig. 1) with an EC_{50} value of approximately 2 μM. Both of the isomers of DOM were able to increase phosphoinositide hydrolysis, the *R*(−)-isomer being more potent and more efficacious than *S*(+)-DOM (table 1). Neither of the isomers of DOM was as efficacious as was 5-HT. The effect of both isomers was blocked by ketanserin, but not by muscarinic, histamine-H1 or *alpha*-1 adrenergic antagonists (fig. 2). Ketanserin and spiperone blocked the effect of racemic DOM (data not shown).

LSD potently stimulated phosphoinositide hydrolysis with an EC_{50} value of 3 nM, but its maximum effect was only about 25% of that produced by 5-HT (fig. 3). Furthermore, the effect of 5-HT was blocked by increasing concentrations of LSD with a maximal blockade equal to the effect of LSD alone (fig. 3). Ketanserin completely blocked the effect of LSD (data not shown).

Effects of LSD and DOM in choroid plexus. Racemic DOM increased phosphoinositide hydrolysis in the choroid plexus in a concentration-dependent manner (fig. 4) with an EC_{50} value of 0.7 μM. The potent 5-HT-1c antagonist, mianserin, was more efficacious than was the less potent 5-HT-1c

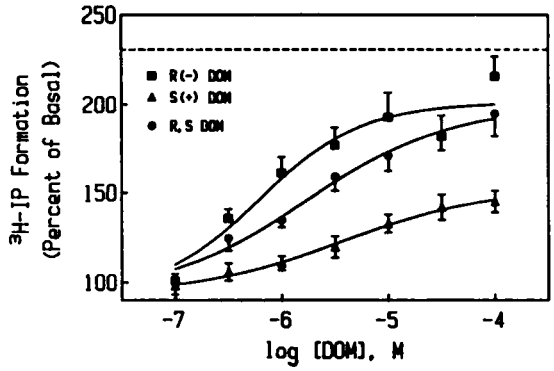

Fig. 1. Stimulation of phosphoinositide hydrolysis by (*R,S*)-DOM and its isomers in rat cerebral cortical slices. [^{3}H]Inositol-labeled slices were incubated for 45 min with 10 mM LiCl, 10 μM pargyline and increasing concentrations of agonist. Radioactivity in the [^{3}H]IP fraction was expressed as percentage of basal and is plotted on the Y axis. The data for racemic DOM reflect two separate experiments, each with an $n = 5$. The data for the isomers of DOM are the mean of 8 to 10 determinations. The vertical bars represent S.E.M. Although the mean basal [^{3}H]IP formation varied from experiment to experiment (the extremes being 701 ± 24 cpm and 204 ± 10 cpm), the fold increase by 5-HT was reproducible. The effect of 100 μM 5-HT is shown by the dashed line (231 ± 7% of basal; $n = 27$).

TABLE 1

Hallucinogen effects at phosphoinositide hydrolysis-linked receptors in cerebral cortex and choroid plexus

The values for 5-HT were taken from Conn *et al.* (1986). The other values were derived from the concentration-response curves presented in figures 1, 3, 4 and 5, using a least square curve fitting program (Graph-Pad, ISI Software). This program calculates EC_{50} values, using the equation for a sigmoid curve and gives estimates of the S.E., which are included in the table. E_{max} values are presented as percentage of the maximum effect of 5-HT analyzed simultaneously.

Agonist	Cortex		Choroid Plexus	
	−log EC_{50}	E_{max}	−log EC_{50}	E_{max}
		% 5-HT		% 5-HT
5-HT	6	100	8.1	100
R,S-DOM	5.8 ± 0.2	76	6.1 ± 0.2	67
R(−)-DOM	6.2 ± 0.2	77	6.5 ± 0.2	91
S(+)-DOM	5.5 ± 0.1	40	5.5 ± 0.2	78
(+)-LSD	8.5 ± 0.1	25	7.9 ± 0.1	34

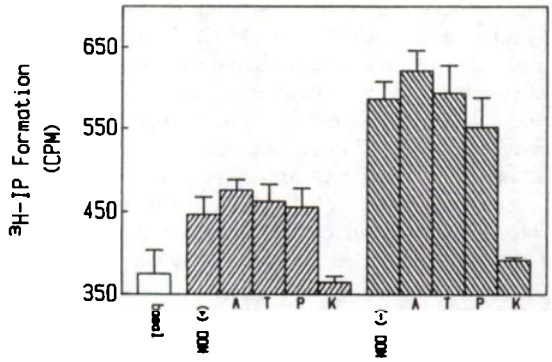

Fig. 2. Antagonism of *S*(+)- and *R*(−)-DOM-stimulated phosphoinositide hydrolysis in slices of cerebral cortex. Conditions were the same as in figure 1. Antagonists were added 15 min before the addition of the agonist (10 μM). The mean basal [^{3}H]IP formation was 375 ± 29 cpm. A, atropine (10 μM); T, triprolidine (10 μM); P, prazosin (10 μM); K, ketanserin (0.32 μM). Vertical bars represent S.E.M.; $n = 4$.

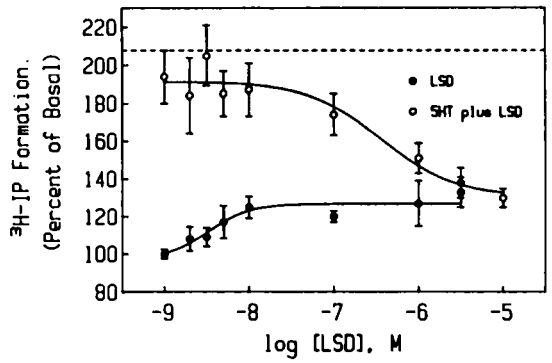

Fig. 3. Partial agonist effect of LSD on 5-HT-2 mediated phosphoinositide hydrolysis in rat cerebral cortex. Conditions were the same as in figure 1. [^{3}H]IP formation is represented as a percentage of the basal radioactivity. The solid circles represent a dose-response curve for LSD-stimulated phosphoinositide hydrolysis. The open circles represent the effect of 10 μM 5-HT plus increasing concentrations of LSD. The dashed line shows the effect of 10 μM 5-HT alone. The mean basal [^{3}H]IP formation was 624 ± 60 cpm. The data are the mean of three separate experiments, each done in triplicate. The vertical bars represent S.E.M.

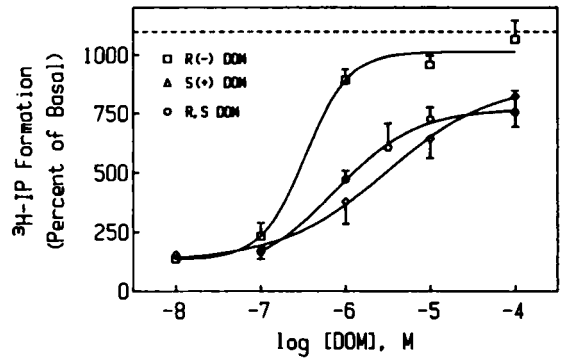

Fig. 4. Effect of (*R*,*S*)-DOM and its isomers on phosphoinositide hydrolysis in choroid plexus. [^{3}H]Inositol-labeled choroid plexi were incubated for 45 min with 10 mM LiCl, 10 μM pargyline and increasing concentrations of agonists (n = 5–6). The mean basal [^{3}H]IP formation ranged from 523 ± 64 cpm to 708 ± 63 cpm. The vertical bars represent S.E.M. The mean effect of 10 μM 5-HT, represented by the dashed line, was 1098% of basal (n = 8).

antagonist spiperone at blocking the effect of *R*,*S*-DOM (data not shown). As in the cerebral cortex, the *R*(−)-isomer of DOM was about 10-fold more potent than the *S*(+)-isomer (table 1).

LSD stimulated phosphoinositide hydrolysis in choroid plexus with an EC_{50} value of approximately 12 nM (fig. 5). Although LSD caused a 4-fold increase in [^{3}H]IP formation, this was still only about 35% of that induced by the full agonist 5-HT. Consistent with a partial agonist effect, increasing concentrations of LSD incompletely antagonized the phosphoinositide hydrolysis response of 5-HT (fig. 5). Mianserin was more effective than spiperone at blocking the effect of LSD (data not shown).

Discussion

These studies show for the first time, using *in vitro* functional correlates of the 5-HT-2 and the 5-HT-1c receptor sites, that the hallucinogens LSD and DOM are direct agonists at these receptors. Agonist properties were evaluated using the phosphoinositide hydrolysis response in isolated tissues. Indirect effects mediated by the release of 5-HT have not been ruled out entirely; however, these seem unlikely to contribute substantially to the effects of the hallucinogens based on the following reasoning: First, although certain amphetamine derivatives have been shown to release 5-HT from brain slices (Hoffman *et al.*, 1986), LSD and DOM have not been reported to promote 5-HT release; indeed, LSD inhibits electrically evoked 5-HT release presumably by activating presynaptic autoreceptors (Farnebo and Hamberger, 1971). Second, the tissues are washed extensively and preincubated for 2 to 3 hr before the addition of the agonists. Because MAO is not inhibited during the preincubation, endogenous 5-HT is at least partially destroyed during this time. Third, the level of 5-HT in choroid plexus is very low (Moskowitz *et al.*, 1979), reducing the likelihood that release mechanisms contribute to hallucinogen-induced phosphoinositide hydrolysis responses in this tissue. Fourth, the effect of LSD is retained in cortical brain slices and in choroid plexi obtained from animals pretreated with the 5-HT neurotoxin, 5,7-dihydroxytryptamine, that

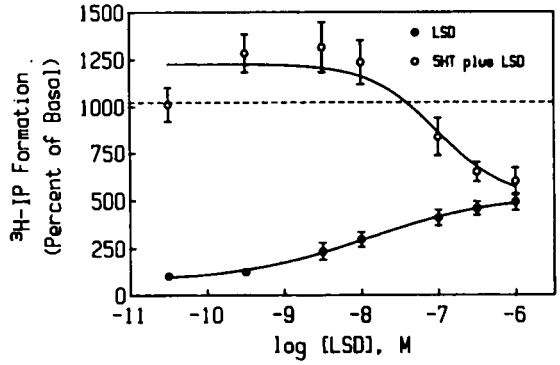

Fig. 5. Partial agonist effect of LSD on 5-HT-1c receptor-mediated phosphoinositide hydrolysis in rat choroid plexus. Conditions were the same as in figure 4. [^{3}H]IP formation is represented as a percentage of the basal radioactivity. The closed circles represent a dose-response curve for LSD alone (n = 4). The open circles show the effect of 100 nM 5-HT plus increasing concentrations of LSD (n = 4). The dashed line shows the effect of 100 nM 5-HT alone (1022 ± 52% of basal). The maximum [^{3}H]IP formation in the presence of 1 μM 5-HT was 1363 ± 143% of basal (n = 4). The mean basal [^{3}H]IP formation was 729 ± 117 cpm (n = 4).

causes >90% depletion of brain 5-HT (unpublished results). It has not been determined if DOM is similarly active in lesioned rats. Taken together, these data make it unlikely that the effects of LSD on phosphoinositide hydrolysis are mediated indirectly by the release of 5-HT. However, the possibility that 5-HT release plays a role in the actions of DOM on phosphoinositide hydrolysis has not been ruled out.

LSD is much more potent than is DOM in stimulating phosphoinositide hydrolysis coupled to 5-HT-2 receptors in cerebral cortex. This finding agrees with behavioral (Aldous *et al.*, 1974; Glennon *et al.*, 1984; Mokler *et al.*, 1985) and electrophysiological (Rasmussen and Aghajanian, 1986) studies in which LSD was demonstrated to be a more potent agonist than DOM. LSD appears to be a partial agonist of the 5-HT-2 receptor-mediated phosphoinositide hydrolysis response, giving a maximum response that is only 25% of that produced by 5-HT, a putative full agonist. The effect of 5-HT is potently, but incompletely, blocked by LSD, as would be predicted for a partial agonist. Furthermore, the effects of 5-HT and LSD on phosphoinositide hydrolysis are not additive, consistent with the interpretation that these agents are acting at the same receptor population. Racemic DOM also stimulates phosphoinositide hydrolysis in the cerebral cortex with a maximum effect that is 76% of that produced by 5-HT. This effect of DOM is mediated apparently by activation of 5-HT-2 receptors, inasmuch as it is blocked by the 5-HT-2 antagonists ketanserin and spiperone, but not by antagonists of other phosphoinositide hydrolysis-linked receptors (*alpha*-1 adrenergic, histaminergic and muscarinic cholinergic). *R*(−)-DOM is the centrally active isomer in humans (Shulgin, 1973), and it is more potent than the *S*(+)-isomer at eliciting smooth muscle contraction (Dyer *et al.*, 1973) and avoidance behavior (Benington *et al.*, 1973). In agreement with these observations, (−)-DOM has a higher potency and greater efficacy than does (+)-DOM for the 5-HT-2 receptor mediating the phosphoinositide response in cerebral cortex.

The effects of LSD and DOM on phosphoinositide hydrolysis in the choroid plexus closely mimic the effects at the cortical 5-HT-2 receptor (table 1). However, the receptor that mediates the phosphoinositide hydrolysis response in choroid plexus is apparently not the 5-HT-2 site. First, there are no 5-HT-2 binding sites in the choroid plexus; instead, 5-HT antagonist radioligands label a novel site, the 5-HT-1c receptor (Pazos *et al.*, 1984; Hoyer *et al.*, 1986; Yagaloff and Hartig, 1985). Second, 5-HT elicits a strong phosphoinositide hydrolysis response in the choroid plexus that is mediated by activation of 5-HT-1c receptor sites, whereas other phosphoinositide hydrolysis-linked receptors are apparently absent in this tissue (Conn *et al.*, 1986; Conn and Sanders-Bush 1986b). Third, the effects of LSD and DOM are blocked by the 5-HT antagonists, mianserin and spiperone, with an order of potency consistent with an interaction with the 5-HT-1c receptor. DOM and LSD both appear to be partial agonists of the 5-HT-1c receptor in choroid plexus, with maximal effects that are less than the maximal effect of 5-HT. LSD is the least efficacious and it potently blocks the effect of 5-HT to a maximum level that is equal to its own intrinsic activity (34% of the maximum effect of 5-HT). The absence of an additive effect of LSD and 5-HT is consistent with the interpretation that LSD and 5-HT interact with the same receptor population in the choroid plexus. As in the cerebral cortex, the *R*(−)-isomer of DOM is more potent than the *S*(+)-isomer in the choroid plexus.

The present demonstration that hallucinogens are agonists at central 5-HT-2 receptors linked to phosphoinositide hydrolysis is consistent with the growing body of behavioral and electrophysiological data that suggest an agonist effect of hallucinogens at 5-HT-2 receptors. However, in some systems, LSD apparently behaves as a 5-HT-2 receptor antagonist; for example, LSD antagonizes 5-hydroxytryptophan-induced head twitches (Gerber *et al.*, 1985) and 5-HT elicited smooth muscle contraction (Van Neuten *et al.*, 1982; Cohen *et al.*, 1985). These apparent contradictory results are in fact not so, considering our finding that LSD is a partial agonist at 5-HT-2 receptors with a low efficacy relative to the full agonist, 5-HT. By definition, a partial agonist with low efficacy should have antagonist properties, because it occupies sites and prevents the binding of a full agonist. Indeed, the functional consequences of the interaction of a low efficacious, partial agonist depends upon the level of ongoing activity within the particular system, *i.e.*, the concentration of the endogenous agonist in the vicinity of the receptor. Thus, depending on the concentration of 5-HT, LSD will have apparently opposite effects on 5-HT-2 receptors, *i.e.*, with a low level of 5-HT, LSD will function as an agonist, because it gives weak, but detectable, receptor stimulation; at higher concentrations of 5-HT, LSD would function as an antagonist because it blocks the effect of 5-HT.

The physiological consequence of hallucinogen interaction at central 5-HT-1c receptors linked to phosphoinositide hydrolysis is unknown. At present, there is no known physiological role for the 5-HT-1c receptors. 5-HT-1c receptors are highly concentrated in the choroid plexus, and we have preliminary evidence that they may function to regulate the composition of the cerebrospinal fluid (Tsutsumi *et al.*, 1988). The finding that hallucinogens (present results) and certain piperazine derivatives (Conn and Sanders-Bush, 1987) function as partial agonists at the 5-HT-1c receptor site may help unravel the mystery of the role of this receptor in choroid plexus function.

In conclusion, 5-HT-2 receptor-mediated phosphoinositide hydrolysis in rat cerebral cortex and the 5-HT-1c receptor-mediated phosphoinositide hydrolysis response in rat choroid plexus have been utilized to determine the direct effects of representative hallucinogens of the indolealkylamine and phenethylamine classes. Whether or not phosphoinositide hydrolysis contributes to the hallucinogenic effects of these drugs remains to be seen. However, agonist-mediated phosphoinositide hydrolysis provides a method by which the potency and efficacy of hallucinogens can be evaluated directly.

Acknowledgments

This work was supported in part by research grants from the National Institute of Mental Health (MH 34007) and from the National Institute of Drug Abuse (DA 05181) and by a training grant from the National Institute of General Medical Sciences (GM 07628).

References

ALDOUS, F. A. B., BARASS, B. C., BREWSTER, K., BUXTON, D. A., GREEN, D. M., PINDER, R. M., RICH, P., SKEELS, M. AND TUTT, K. J.: Structure-activity relationships in psychotomimetic phenylalkylamines. J. Med. Chem. **17**: 1100–1111, 1974.

BENINGTON, F., MORIN, R. D., BEATON, J., SMYTHIES, J. R. AND BRADLEY, R. J.: Comparative effects of stereoisomers of hallucinogenic amphetamines. Nat. New Biol. **242**: 185–186, 1973.

BERRIDGE, M. J., DOWNES, P. C. AND HANLEY, M. R.: Lithium amplifies agonist-dependent phosphatidylinositol responses in brain and salivary glands. Biochem. J. **206**: 587–595, 1982.

COLPAERT, F. C., MEERT, T. F., NIEMEGEERS, C. J. E. AND JANSSEN, P. A. J.: Behavioral and 5-HT antagonist effects of ritanserin: A pure and selective antagonist of LSD discrimination in rat. Psychopharmacology **86**: 45–54, 1985.

COHEN, M. L., SCHENCK, K. W., COLBERT, W. AND WITTENAUER, L.: Role of 5-

HT-2 receptors in serotonin-induced contractions of nonvascular smooth muscle. J. Pharmacol. Exp. Ther. **232:** 770–774, 1985.

CONN, P. J. AND SANDERS-BUSH, E.: Agonist-induced phosphoinositide hydrolysis in choroid plexus. J. Neurochem. **47:** 1754–1760, 1986b.

CONN, P. J. AND SANDERS-BUSH, E.: Regulation of serotonin-stimulated phosphoinositide hydrolysis: Relation to the serotonin 5-HT-2 binding site. J. Neurosci. **6:** 3669–3675, 1986a.

CONN, P. J. AND SANDERS-BUSH, E.: Serotonin-stimulated phosphoinositide turnover: Mediation by the S_2 binding site in rat cerebral cortex but not in subcortical regions. J. Pharmacol. Exp. Ther. **234:** 195–203, 1985.

CONN, P. J. AND SANDERS-BUSH, E.: Relative efficacies of piperazines at the phosphoinositide hydrolysis-linked serotonergic (5-HT-2 and 5-HT-1c) receptors. J. Pharmacol. Exp. Ther. **242:** 552–557, 1987.

CONN, P. J. AND SANDERS-BUSH, E.: Selective 5-HT-2 antagonists inhibit serotonin stimulated phosphatidylinositol metabolism in cerebral cortex. Neuropharmacology **23:** 993–996, 1984.

CONN, P. J., SANDERS-BUSH, E., HOFFMAN, B. J. AND HARTIG, P. R.: A unique serotonin receptor in choroid plexus is linked to phosphatidylinositol turnover. Proc. Natl. Acad. Sci. U.S.A. **83:** 4086–4088, 1986.

DYER, D. C., NICHOLS, D. E., RESTERHOLZ, D. AND BARFKNECHT, C.F.: Comparative effects of stereoisomers of psychotomimetic phenylisopropylamines. Life Sci. **13:** 885–896, 1973.

ENGEL, G., GOTHERT, M., HOYER, D., SCHLICKER, E. AND HILLENBRAND, K.: Identity of inhibitory presynaptic 5-hydroxytryptamine (5HT) autoreceptors in the rat hrain cortex with 5HT-1b binding sites. Naunyn-Schmiedeberg's Arch. Pharmacol. **332:** 1–7, 1986.

FARNEBO, L. O. AND HAMBERGER, B.: Drug-induced changes in the release of [^{3}H]-monoamines from field stimulated rat brain slices. Acta Physiol. Scand. **371:** 35–44, 1971.

GERBER, R., BARBAZ, B. J., MARTIN, L. L., NEALE, R., WILLIAMS, M. AND LIEBMAN, J. M.: Antagonism of L-5-hydroxytryptophan-induced head twitching by lisuride: A mixed 5-hydroxytryptamine agonist-antagonist? Neurosci. Lett. **60:** 207–213, 1985.

GLENNON, R. A., TITELER, M. AND MCKENNEY, J. D.: Evidence for 5-HT_2 involvement in the mechanism of action of hallucinogenic agents. Life Sci. **35:** 2505–2511, 1984.

HOFFMAN, A. J., MAXWELL, D. M. AND NICHOLS, D. E.: A simple organ bath for electrical stimulation and superfusion of rat brain slices. J. Pharmacol. Methods **16:** 185–199, 1986.

HOYER, D., SRIVATSA, S., PAZOS, A., ENGEL, G. AND PALACIOS, J. M.: [^{125}I]LSD labels 5-HT_{1C} recognition sites in pig choroid plexus membranes. Comparison with [^{3}H]mesulergine and [^{3}H]5-HT binding. Neurosci. Lett. **69:** 269–274, 1986.

MOKLER, D. J., STOUDT, K. W. AND RECH, R. H.: The 5HT2 antagonist pirenperone reverses diaruption of FR-40 by hallucinogenic drugs. Pharmacol. Biochem. Behav. **22:** 677–682, 1985.

MOSKOWITZ, M. A., LIEBMANN, J. E., REINHARD, J. F., JR. AND SCHLOSBERG, A.: Raphe origin of serotonin-containing neurons within choroid plexus of the rat. Brain Res. **169:** 590–594, 1979.

PAZOS, A., HOYER, D. AND PALACIOS, J. M.: The binding of serotonergic ligand to the porcine choroid plexus: Characterization of a new type of serotonin recognition site. Eur. J. Pharmacol. **106:** 539–546, 1984.

PENINGTON, N. J. AND REIFFENSTEIN, R. J.: Possible involvement of serotonin receptors in the facilitatory effect of a hallucinogenic phenethylamine on single facial motoneurons. Can. J. Physiol. Pharmacol. **64:** 1302–1309, 1986.

PEROUTKA, S. J.: Pharmacological differentiation and characterization of 5HT1a, 5HT1b and 5HT1c binding sites in rat frontal cortex. J. Neurochem. **47:** 529–540, 1986.

RASMUSSEN, K. AND AGHAJANIAN, G. K.: Effect of hallucinogens on spontaneous and sensory-evoked locus coeruleus unit activity in the rat: Reversal by selective 5-HT_2 antagonists. Brain Res. **385:** 395–400, 1986.

RASMUSSEN, K., GLENNON, R. A. AND AGHAJANIAN, G. K.: Phenethylamine hallucinogens in the locus coeruleus: Potency of action correlates with rank order of 5-HT_2 binding affinities. Eur. J. Pharmacol. **132:** 79–82, 1986.

SANDERS-BUSH, E. AND CONN, P. J.: Neurochemistry of serotonin neuronal systems: Consequences of serotonin receptor activation. *In* Psychopharmacology: The Third Generation of Progress, ed. by H. Meltzer, pp. 95–103, Raven Press, New York 1987.

SHULGIN, A. T.: Stereospecific requirements for hallucinogenesis. J. Pharm. Pharmacol. **25:** 271–272, 1973.

TSUTSUMI, M., SKINNER, M. AND SANDERS-BUSH, E.: Transferrin secretion by rat choroid plexus is modulated by serotonin and insulin. FASEB J. **2:** A1404, 1988.

VAN NEUTEN, J. M., LEYSEN, J. E. VANHOUTTE, P. M. AND JANSSEN, P. A. J.: Serotonergic responses in vascular and nonvascular tissues. Arch. Int. Pharmacodyn. **256:** 331–334, 1982.

YAGALOFF, K. A. AND HARTIG, P. R.: ^{125}I-Lysergic acid diethylamide binds to a novel serotonergic site on rat choroid plexus epithelial cells. J. Neurosci. **5:** 3178–3183, 1985.

Send reprint requests to: Elaine Sanders-Bush, Ph.D., Department of Pharmacology, A5321 Medical Center North, Vanderbilt University School of Medicine, Nashville, TN 37232.