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Effects of 5-HT_{1A} Receptor Agonists and L-5-HTP in Montgomery's Conflict Test

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SÖDERPALM, B., S. HJORTH AND J. A. ENGEL. *Effects of 5-HT_{1A} receptor agonists and L-5-HTP in Montgomery's conflict test.* PHARMACOL BIOCHEM BEHAV 32(1) 259–265, 1989.—The effects of the pyrimidinyl-piperazines buspirone, gepirone, ipsapirone and their common metabolite 1-(2-pyrimidinyl)-piperazine (PmP) as well as of 8-hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT) and L-5-hydroxytryptophan (L-5-HTP) were investigated in Montgomery's conflict test—an animal anxiety model based on the animal's inborn urge to explore a new environment and its simultaneous fear of elevated, open spaces. Subcutaneous buspirone (32–128 nmol/kg), gepirone (32–128 nmol/kg), ipsapirone (32–512 nmol/kg) and 8-OH-DPAT (50–200 nmol/kg), as well as intraperitoneal L-5-HTP (56 μmol/kg) produced anxiolytic-like effects. However, at higher doses the magnitude of these effects decreased and overall the dose-response curves displayed inverted U-shapes. The highest doses (2048 nmol/kg) of buspirone and of gepirone even decreased responding below control levels, possibly in part due to concomitant sedation/motor impairment. After L-5-HTP (448 μmol/kg) and PmP (512 nmol/kg) anxiogenic-like effects were observed. The results indicate that anxiolytic- and anxiogenic-like effects of drugs affecting central serotonergic neurotransmission can be obtained in a sensitive rat anxiety model which neither involves consummatory behavior nor punishment. The anxiolytic-like effects of these compounds may be due to their 5-HT_{1A} agonistic properties. Moreover, the present data may provide support for a possible reciprocal association of presynaptic 5-HT_{1A} receptors vs. postsynaptic 5-HT_{1A} as well as 5-HT_2 receptors with regard to anxiety.

Anxiolytic-like effect Anxiogenic-like effect Buspirone 8-OH-DPAT 5-HT_{1A} receptors Gepirone Ipsapirone L-5-HTP Montgomery's conflict test PmP Rat Serotonin

DRUGS affecting brain serotonergic (5-HT) neurons have lately attracted increasing interest as putative anxiolytic compounds. For example, both the 5-HT_{1A} receptor agonist buspirone and the 5-HT_2 receptor antagonist ritanserin have been reported effective in the treatment of generalized anxiety (7,46). Moreover, recent clinical studies indicate that the 5-HT precursor L-5-hydroxytryptophan (L-5-HTP) may be effective in the treatment both of generalized anxiety and panic anxiety (27,28). The latter disorder is also highly susceptible to treatment with monoamine oxidase inhibitors and tricyclic antidepressants [for a review, see (35)], drugs which have marked effects on brain 5-HT systems. The anxiolytics most widely used, the benzodiazepines (BDZs), also influence 5-HT neurotransmission. Whether this action is related to the anxiolytic effects of these drugs is, however, controversial [cf. (29, 44, 47)].

In animal models believed to reflect anxiety-related mechanisms, manipulations resulting in decreased and increased brain serotonergic activity may lead to anxiolytic- and anxiogenic-like actions, respectively. For example, depletion of 5-HT by means of the tryptophan hydroxylase inhibitor *para*-chlorophenylalanine (PCPA), or selective destruction of 5-HT neurons by means of 5,7-dihydroxytryptamine (5,7-DHT), have repeatedly been shown to produce anxiolytic-like effects in reward/punishment conflict procedures [cf. (29,44)]. Moreover, L-5-HTP induced, depending on the dose applied, either anxiolytic- or anxiogenic-like effects in a modified Vogel's drinking conflict test (25). Some investigators report that 5-HT_{1A} receptor agonists produce anticonflict (anxiolytic-like) effects, while others have failed to find such effects or even observed anxiogenic-like actions of these compounds [for references, see (13)].

Brain 5-HT neurons have been implicated also in the control of food and liquid intake was well as in pain sensitivity (3, 8, 40). Many animal anxiety models comprise feeding or drinking drives paired with aversive stimulation (e.g., electrical shock). Even though putative drug effects on these drives and/or on shock sensitivity have been controlled for in a few studies, it is desirable to determine if anxiolytic-like actions of 5-HT active drugs can be established also in models devoid of such potentially confounding factors. We have therefore examined the effects of different putative 5-HT_{1A} receptor agonists and of L-5-HTP in Montgomery's conflict test, an animal anxiety model involving neither consummatory behavior nor punishment.

METHOD

Male Sprague-Dawley rats (ALAB, Stockholm, Sweden) weighing 250–350 g were used. The animals were kept under

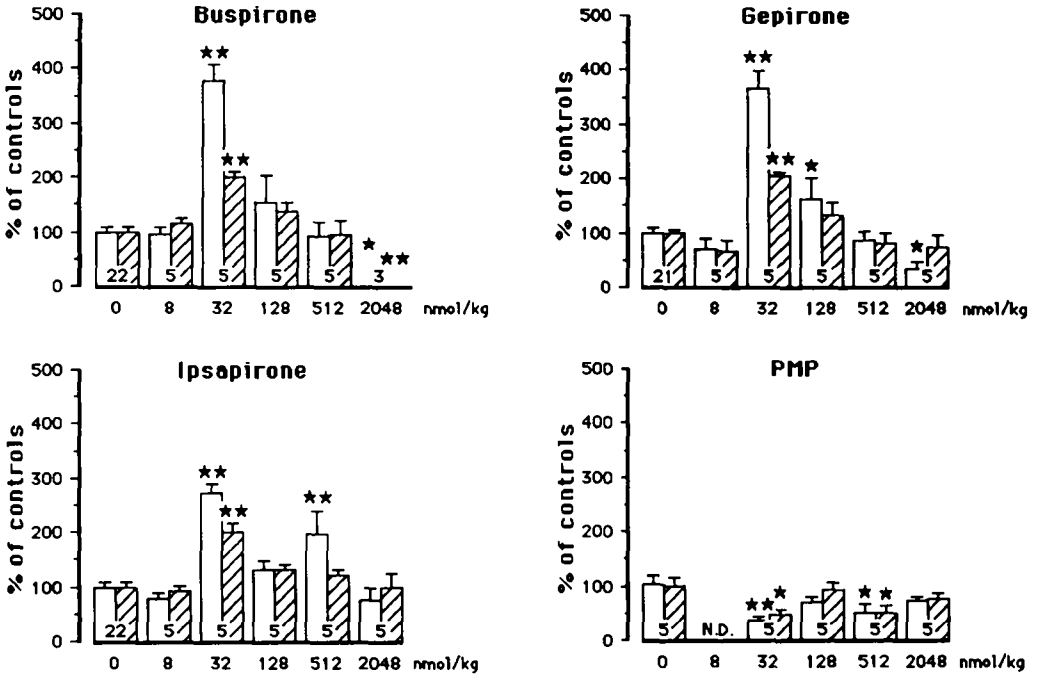

FIG. 1. Effects of piperazine-pyrimidinyl derivatives in Montgomery's conflict test. Drugs were injected SC 10 min prior to the test. Eight nmol of buspirone, gepirone and ipsapirone equals 3.0–3.5 μg (HCl), 8 nmol of PmP equals 1.7 μg (HCl). The recorded data were treated as outlined under the Method section and are presented as percent of controls. Shown are the means±S.E.M. of 3–22 observations. Open columns=time spent in open arms; filled columns=entries made into open arms. N.D.=not determined. Statistics: ANOVA, buspirone: time, $F(5,39)=25.47$, $p<0.01$; entries, $F(5,39)=12.55$, $p<0.01$, gepirone: time, $F(5,40)=31.97$, $p<0.01$; entries, $F(5,40)=11.85$, $p<0.01$, ipsapirone: time, $F(5,41)=13.17$, $p<0.01$; entries, $F(5,41)=7.20$, $p<0.01$, PmP: time, $F(4,20)=4.06$, $p<0.05$; entries, $F(4,20)=3.58$, $p<0.05$, followed by Dunnett's t-test: ★$p<0.05$, ★★$p<0.01$.

controlled light-dark conditions (lights on at 5.00 a.m. and off at 7.00 p.m.) and at constant temperature (25°C) and humidity (65%). An adaptation period of at least seven days to the animal maintenance facilities of the Department was allowed prior to the start of the experiments. The animals had free access to standard laboratory chow and tap water.

Montgomery's Conflict Test

The experimental device was an elevated (1 m above the ground), +-shaped maze, which was placed in a dimly lit room. The four arms were 40 cm long and 10 cm wide, and had wire-mesh floors. Two opposing arms were surrounded by black, 10 cm high Plexiglas walls (closed arms), while the other arms were devoid of walls (open arms). The floor underneath the maze was covered with bright white paper.

The animal was initially removed from its home cage and put in an unfamiliar environment for five minutes. Thereafter it was placed in the center of the maze, facing a closed arm. The observer was situated 2 m from the center of the maze. Entry into an arm was defined as the animal placing all four paws onto the arm. The cumulative time spent in, and the number of entries made into, the open or closed arms, were recorded during a five-minute test-session. The open arm data were then expressed as percent of the total time spent in, and of the total number of entries made into, both open and closed arms.

The plus-maze was carefully wiped with a wet towel after each tested animal. All experiments were carried out between 10 a.m. and 4 p.m. During this period control rats respond uniformly (Söderpalm and Engel, unpublished data). All animals were drug-naive and used only once. Introduction of the animal to an unfamiliar environment prior to the test increases the total number of entries [(37); Söderpalm and Engel, unpublished data], thus reducing the risk of erroneous random distribution of the open/closed arm ratios.

Well-known anxiolytics, such as diazepam, increase the time spent in, and the number of entries made into open arms. Conversely, anxiogenic compounds reduce both of these measures (38). On the other hand, drugs with sedative properties but without specific anxiolytic action, have no effects on the open/closed arm relationships (37). The conflict state in Montgomery's maze is believed to be derived from the animal's inborn urge to explore the new environment and its simultaneous fear of elevated, open spaces.

Drugs

The drugs used in the studies were: buspirone (MJ 9022-1 [8-(4,4-(2-pyrimidinyl)-1-piperazinyl(butyl)-8-azaspiro[4.5] decane-7,9-dione × HCl]), gepirone (MJ 13805 [4,4-dimethyl-1-(4-(2-pyrimidinyl)-1-piperazinyl)-butyl)-2,6-piper-

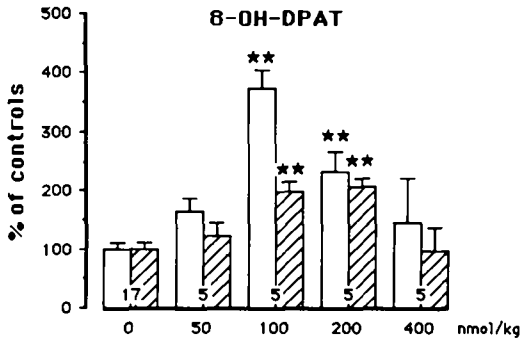

FIG. 2. Effects of 8-OH-DPAT in Montgomery's conflict test. The drug was injected SC 10 min prior to the test. Fifty nmol of 8-OH-DPAT equals 16 μg (HBr). The recorded data were treated as outlined under the Method section and are presented as percent of controls. Shown are the means±S.E.M. off 5–17 observations. Open columns=time spent in open arms; filled columns=entries made into open arms. Statistics: ANOVA, time, $F(4,32)=12.11$, $p<0.01$; entries, $F(4,32)=8.31$, $p<0.01$, followed by Dunnett's t-test: ★★$p<0.01$.

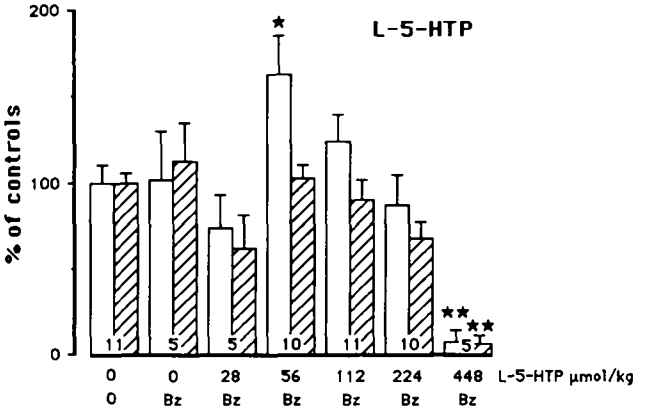

FIG. 3. Effects of L-5-HTP and the peripheral decarboxylase inhibitor benserazide 25 mg/kg (Bz) in Montgomery's conflict test. Bz and L-5-HTP were administered IP 60 and 30 min, respectively, prior to the test. Twenty-eight μmol of L-5-HTP equals 6.25 mg (base). The recorded data were treated as outlined under the Method section and are presented as percent of controls. Shown are the means±S.E.M. of 5–11 observations. Open columns=time spent in open arms; filled columns=entries made into open arms. Statistics: ANOVA, time, $F(6,50)=5.69$, $p<0.01$; entries, $F(6,50)=7.63$, $p<0.01$, followed by Dunnett's t-test: ★$p<0.05$, ★★$p<0.01$.

idinedione × HCl]), PmP (MJ 13653 [1-(2-pyrimidinyl)piperazine × HCl]) (all three agents: courtesy Dr. D. P. Taylor, Bristol-Myers Company, Wallingford, CT), ipsapirone (TVX Q 7821: [2-(4-(4-(2-pyrimidinyl)-1-(piperazinyl)butyl-1,2-benzisothiazol-3-[2H]one-1,1-dioxide × HCl]; courtesy Dr. J. Traber, Troponwerke, Cologne, FRG), 8-hydroxy-2(di-*n*-propyl-amino)tetralin (8-OH-DPAT × HBr; courtesy Merck Sharp & Dohme, West Point, PA), L-5-hydroxytryptophan (L-5-HTP; Sigma, St. Louis, MO) and benserazide (Ro 4-4602/1; courtesy Hoffmann-La Roche, Basle, Switzerland). L-5-HTP was dissolved in a minimal quantity of 1 N HCl and made up to volume with 0.9% saline. All other compounds were dissolved in physiological saline only. Benserazide and L-5-HTP were given IP at 60 and 30 min, respectively, before the test session. The other drugs were injected SC 10 min prior to placing the animals in the maze. Injection volumes were 2 ml/kg (benserazide) or 5 ml/kg (other drugs). The doses are given in nmol or μmol/kg; 8 nmol of buspirone, gepirone and ipsapirone equals 3.0–3.5 μg (HCl), 8 nmol of PmP equals 1.7 μg (HCl), 50 nmol of 8-OH-DPAT equals 16 μg (HBR), and 28 μmol of L-5-HTP equals 6.25 mg (base).

Statistics

Differences between treatment groups were statistically evaluated by means of one-way analysis of variance (ANOVA) followed by Dunnett's t-test. Probabilities of less than 5% were considered significant.

RESULTS

The total number of entries (open + closed arm entries) made by control rats during the five-minute test-session was about 13–14 in the present set of experiments. Twenty to 30% of the entries were made into the open arms (OE), and 7.5–17.5% of the total time (190–240 sec) spent in arms (any type) was spent in the open arms (OT).

The pyrimidinyl-piperazine derivatives buspirone (32–128 nmol/kg), gepirone (32–128 nmol/kg) and ipsapirone (32–512 nmol/kg) all increased, as compared to controls, OE and/or OT in Montgomery's test (Fig. 1). At the highest dose tried (2048 nmol/kg), buspirone instead significantly decreased both of these measures, while this dose of gepirone significantly lowered the time parameter (OT) only (Fig. 1). The pyrimidinyl-piperazine metabolite PmP generally tended to decrease both OE and OT; these changes were significant, compared to controls, at 32 and 512 nmol/kg (Fig. 1).

8-OH-DPAT increased OE as well as OT (Fig. 2). However, similarly to buspirone, gepirone and ipsapirone, the 8-OH-DPAT dose-response curve displayed an inverted U-shape; increases in OE and OT at low (50 nmol/kg; not significant) and intermediate doses (100–200 nmol/kg; $p<0.01$), but no effect at 400 nmol/kg.

L-5-HTP (after peripheral decarboxylase inhibition by means of benserazide 25 mg/kg) produced a biphasic dose-response curve in Montgomery's test (Fig. 3). A single low dose (56 μmol/kg) significantly increased OT, whereas the highest dose tried (448 μmol/kg) instead caused highly significant decreases with respect both to OE and OT. Indeed, judging from gross behavioral observations, these latter animals gave the impression of being very apprehensive towards entering the open arms. Benserazide per se did not affect the behavior.

The total number of entries (i.e., open + closed arm entries/test session) was used as an index of drug effects on overall motor activity. As seen in Table 1, the total number of entries made during the five-minute test period was, by and large, unaffected by most drugs and doses. The lowest dose of buspirone (8 nmol/kg) slightly, though significantly, increased the total number of entries. However, the highest doses (2048 nmol/kg) of buspirone, gepirone and ipsapirone as well as of 8-OH-DPAT (400 nmol/kg), instead clearly decreased this measure. Buspirone exerted the most, and ipsapirone the least marked effect in this context. L-5-HTP

TABLE 1

EFFECTS ON LOCOMOTOR ACTIVITY IN MONTGOMERY'S CONFLICT TEST

	0	8	32	128	512	2048
			nmol/kg			
Buspirone	13.6 ± 0.6	18.0 ± 1.7*	16.0 ± 1.1	15.2 ± 1.2	12.0 ± 2.1	2.7 ± 0.3†
Gepirone	13.8 ± 0.6	15.6 ± 1.3	15.4 ± 0.5	11.8 ± 1.8	10.8 ± 0.8	6.6 ± 1.6†
Ipsapirone	13.6 ± 0.6	16.4 ± 1.3	15.4 ± 1.0	14.6 ± 1.0	12.8 ± 1.1	9.8 ± 1.4*
PmP	15.2 ± 1.0	N.D.	11.4 ± 0.9	18.2 ± 1.6	13.4 ± 0.8	13.6 ± 1.5
	0	50	100	200	400	
			nmol/kg			
8-OH-DPAT	13.1 ± 0.7	15.2 ± 0.7	14.8 ± 1.4	11.0 ± 1.6	4.0 ± 0.7†	
	0	28	56	112	224	448
			μmol/kg			
L-5-HTP	13.0 ± 0.7	11.2 ± 1.7	15.4 ± 0.9	16.0 ± 0.9	17.0 ± 1.3*	14.0 ± 1.6

Drug effects on the total number of entries made during five minutes in Montgomery's conflict test. L-5-HTP was injected IP 30 min prior to the test. The other drugs were injected SC 10 min prior to the test. Eight nmol of buspirone, gepirone and ipsapirone equals 3.0–3.5 μg (HCl), 8 nmol of PmP equals 1.7 μg (HCl), 50 nmol of 8-OH-DPAT equals 16 μg (HBr), and 28 μmol of L-5-HTP equals 6.25 mg (base). Shown are the means ± S.E.M. N.D.=not determined. Statistics: ANOVA, buspirone, $F(5,39)=10.55$, $p<0.01$; gepirone, $F(5,40)=8.23$, $p<0.01$; ipsapirone, $F(5,41)=3.58$, $p<0.01$; PmP, $F(4,20)=4.49$, $p<0.01$; 8-OH-DPAT, $F(4,32)=13.76$, $p<0.01$; L-5-HTP, $F(6,50)=2.72$, $p<0.05$, followed by Dunnett's t-test: *$p<0.05$, †$p<0.01$.

(224 μmol/kg), on the other hand, caused a small but significant increase in overall activity. Gross behavioral observations during the tests indicated that sedation and/or apparent motor incapacitation (e.g., flattened body posture) may account for the observed reductions in this index of motor activity, although it cannot be excluded that other effects, e.g., increased anxiety/fear contributed to these results.

DISCUSSION

Buspirone, gepirone, ipsapirone, 8-OH-DPAT and L-5-HTP all, at low doses, induced behavioral changes similar to those seen after administration of well-established anxiolytics (37,43) and, thus, exerted anxiolytic-like effects in Montgomery's conflict test. In these doses none of the drugs significantly altered the total number of entries made into the arms of the maze. Hence, unspecific stimulatory or sedative drug effects are not likely to explain the results obtained. The results are in agreement with earlier reports of anxiolytic-like effects of the compounds listed, in rat in different punished conflict procedures [cf. (15, 25, 52)]. In contrast, during the course of these studies, Pellow *et al.* (39) and Critchley and Handley (9) reported of either anxiogenic-like or lack of effects of 8-OH-DPAT, buspirone and ipsapirone in the elevated plus-maze. The investigators used, however, generally higher doses than those applied in the present study. As suggested by our findings (vide infra), such high doses would not be expected to produce anxiolytic-like effects. Differences with respect to experimental design, including the strains of rats used, are other possible explanations to the discrepant results.

Interestingly, after higher doses of the pyrimidinylpiperazines (128–512 nmol/kg) and 8-OH-DPAT (200 nmol/kg), the open arm time (OT) and entries (OE) tended to return towards control levels. The possibility that this is a result merely reflecting gross motor impairment ("5-HT syndrome" or other) seems, however, less likely since the total number of entries was generally unaffected at such doses. Still higher doses of buspirone and gepirone (2048 nmol/kg), reduced OT or OE *below* control levels. However, at these doses the total number of entries *was* decreased for both drugs (as well as for ipsapirone), thus making the interpretation of the decreases in OT and OE as reflecting anxiogenic-like effects uncertain.

Contrasting to the above, L-5-HTP at the highest dose tested markedly decreased both OT and OE without affecting locomotor activity. Consequently, these data may be interpreted as representing an anxiogenic-like action of L-5-HTP. This idea is further substantiated by the gross behavior displayed by these animals, i.e., a marked apprehension towards the open arms. In general, the L-5-HTP data are in line with results previously obtained in a modified Vogel's drinking conflict test (25); anxiolytic-like effect in a low dose (cf. above) and anxiogenic-like effects in higher doses. The data discussed in this and the former paragraph also illustrate that there is no clearcut correlation between impaired motor function and decreased responding in Montgomery's conflict test.

The present study shows that both anxiolytic- and anxiogenic-like effects of drugs affecting 5-HT neurotransmission can be detected also in an animal anxiety model devoid of drinking and feeding drives paired with aversive stimuli. Moreover, the findings that buspirone (46) and L-5-HTP (27,28) possess anxiolytic properties in humans strengthen the usefulness of Montgomery's test for studying putative anxiolytic compounds. Notably, the doses of 5-HT active agents, and BDZs (43), producing anxiolytic-like effects are generally lower than those required in punished

conflict procedures, indicating a high degree of sensitivity of this test.

Several possibilities are conceivable as regards putative mechanisms involved in the anxiety-related effects described. Even though buspirone has been claimed to alter in vivo BDZ-receptor binding (36), the specificity of this effect remains obscure [see (19)]. In vitro the presently used compounds show no affinity for BDZ-receptors [cf. (48)]. Apart from producing anxiolytic-like effects the behavioral profiles of these drugs also differ from those of BDZs [cf. (48)]. Moreover, diazepam-stimulus generalizations do not occur for these compounds (51). Taken together, these findings make it less likely that the anxiolytic-like effects of the pyrimidinyl-piperazines, 8-OH-DPAT and L-5-HTP are exerted through direct interaction with the BDZ/GABA receptor complex.

Attenuating central dopaminergic transmission can result in anxiolytic-like effects in a punished conflict procedure [cf. (24)]. However, in contrast to buspirone (22,30), neither 8-OH-DPAT (23), gepirone [(30); Hjorth, unpublished] nor ipsapirone [(32); Hjorth, unpublished] appear to possess any prominent DA antagonistic properties. While it might thus be suggested that the anxiolytic-like profile of buspirone stems from its antidopaminergic action, such an interpretation appears less likely to account for the anticonflict effects of the other agents—particularly given the very low doses required.

Like the prototype 5-HT_{1A} agonist 8-OH-DPAT, buspirone, gepirone as well as ipsapirone possess high affinity and selectivity for brain 5-HT_{1A} receptor binding sites [cf. (48)]. Functionally, a wealth of data demonstrates that buspirone, gepirone, ipsapirone and 8-OH-DPAT inhibit rat brain 5-HT neuronal activity [cell firing, 5-HT synthesis, turnover, release; (12, 22, 23, 31, 33), see (48)]. All these effects are compatible with presynaptic 5-HT_{1A} receptor activation. Moreover, to varying extents the drugs produce the so-called 5-HT syndrome, which at least partly is mediated via postsynaptic 5-HT_{1A} receptors (14, 41, 49). There is also electrophysiological evidence, as well as a study on adenylate cyclase activity, to suggest that these compounds can activate postsynaptic 5-HT_{1A} receptors in the rat hippocampus (1, 2, 4, 34). In drug discrimination studies, buspirone, ipsapirone and 8-OH-DPAT readily substitute for each other (11, 45, 49). The latter findings in particular provide strong support for the contention that in vivo, in awake animals, these drugs affect the organism through similar mechanisms. Common denominators for buspirone, gepirone, ipsapirone and 8-OH-DPAT thus appear to be their high affinities for, and their apparent agonistic properties at, brain 5-HT_{1A} receptors. Available data are thus consistent with the idea that the anxiolytic-like effects of these compounds are produced through activation of brain 5-HT_{1A} receptors. Due to the lack of specific 5-HT_{1A} receptor antagonists this issue cannot, however, be firmly established at this point.

It is not clear whether the 5-HT_{1A} receptors putatively involved in the anxiolytic-like effects are pre- or postsynaptic, nor in what brian regions the effects are exerted. Previous animal experiments indicate that decreased 5-HT neurotransmission produces anxiolytic-like effects [cf. (26,29)]. We have recently found that depletion of brain 5-HT by means of PCPA results in anxiolytic-like effects also in Montgomery's conflict test (Söderpalm, Carlsson and Engel, unpublished data). As activation of 5-HT_{1A} receptors on 5-HT cell bodies in the raphe nuclei reduces the electrical activity in these cells, the anxiolytic-like effects of the 5-HT_{1A} receptor agonist may result from a reduction in 5-HT neurotransmission. The net mechanism of action would then resemble that presumed for PCPA. Interestingly, the doses of buspirone exerting anxiolytic-like effects in the present study are similar to those yielding decreased electrical activity in the dorsal raphe (51). Moreover, two recent studies indicate that local application of 5-HT_{1A} receptor agonists in the raphe nuclei elicits anxiolytic-like effects (6,21).

Notably, an earlier report from this laboratory showed that the anticonflict effect of 8-OH-DPAT turned into a "pro"-conflict (anxiogenic-like) effect after pretreatment with PCPA, tentatively suggested to be mediated through the activation of (sensitized?) postsynaptic 5-HT_{1A} receptors (15). The typical shapes of the dose-response cuves (inverted U-shape) obtained in the present study could similarly result from a delicate balance between activation of pre- and postsynaptic 5-HT_{1A} receptors in anxiety-related neuronal circuits. The dose/effect separation might then be due to the higher sensitivity often ascribed the former receptors (autoreceptors) as compared to postsynaptic 5-HT_{1A} receptors, thereby possibly explaining the narrow "anxiolytic-like dose-window" of 5-HT_{1A} agonists.

As mentioned above, unlike the 5-HT_{1A} receptor agonists, L-5-HTP at the highest dose used produced clear-cut anxiogenic-like effects. Since L-5-HTP after transformation to 5-HT would be expected to stimulate all putative types of 5-HT receptors, this could implicate that, in addition to 5-HT_{1A}, other 5-HT receptors (e.g., 5-HT_2 receptors) might be involved in anxiety mechanisms. Ritanserin is an allegedly 5-HT_2-receptor selective antagonist which can produce anxiolytic-like effects in Montgomery's conflict test (9), and which has proven active as an anxiolytic in humans (7). It may be of interest in this context that anxiety, and even panic reactions, may occur after intake of hallucinogenic drugs—the major effect of which has been ascribed their 5-HT_2 receptor agonistic properties (18).

To summarize, the present set of data may provide further support for a possible reciprocal association of presynaptic 5-HT_{1A} receptors vs. postsynaptic 5-HT_{1A} as well as 5-HT_2 receptors with regard to anxiety (25). Needless to say, confirmation of this hypothesis has to await behavioral studies applying microinjections of specific 5-HT receptor agonists and antagonists into various 5-HT cell body and target areas, respectively.

In contrast to the above drugs, PmP failed to produce anxiolytic-like effects. In fact, this compound instead significantly decreased the open arm visits. PmP has been reported to be a potent α_2-adrenoceptor antagonist with only weak 5-HT_1 receptor affinity (5,17). Indeed, its profile in Montgomery's test resembles those previously obtained for α_2-adrenolytics (20,42). An influence of the metabolite PmP could conceivably also underlie the decline in anxiolytic-like effect seen with increasing doses of the parent pyrimidinyl-piperazines. However, in this case, any major influence of PmP—formed in the liver (16)—appears unlikely given the short injection-test interval used (10 min), and the route of administration (SC). Moreover, also the nonpyrimidinyl-piperazines 8-OH-DPAT and L-5-HTP produced inverted U-shaped and biphasic dose-response curves, respectively.

The role of 5-HT in anxiolysis has recently been challenged in a comprehensive review by Soubrié (44). He suggested that the anticonflict effects seen after manipulations of the 5-HT system in rat may be due to an incapacity to withhold responses for an anticipated reward rather than to reduced anxiety. The present investigation indicates,

however, that anxiolytic-like effects after manipulations of 5-HT neurotransmission can be obtained in an anxiety model without such a reward. Furthermore, even though a loss of inhibition of on-going behavior could explain the results obtained in some animal behavioral models, including traditional conflict models, this does not preclude that an anxiolytic effect is concomitantly at hand. As clinical evidence is accumulating for an involvement of 5-HT in anxiety syndromes [for references see Introduction, see also (10)], it seems premature to discard this transmitter system as a target for development of future anxiolytic drugs. On the contrary, the present study suggests that further clinical trials are warranted, with specific 5-HT_{1A} receptor agonists like 8-OH-DPAT, gepirone and ipsapirone in anxiety states.

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