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Inhibition of Mouse Killing Behavior by Serotonin-Mimetic Drugs: Effects of Partial Alterations of Serotonin Neurotransmission

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MOLINA, V., L. CIESIELSKI, S. GOBAILLE, F. ISEL AND P. MANDEL. *Inhibition of mouse killing behavior by serotonin-mimetic drugs: Effects of partial alterations of serotonin neurotransmission.* PHARMACOL BIOCHEM BEHAV **27**(1) 123–131, 1987.—Rats which do not kill mice and which acquire mouse killing behavior after partial lesion of the serotonin neurotransmission, either by p-chlorophenylalanine treatment or by electrolytical lesions of dorsal and median raphe nucleus, were treated by IP injection of serotonin-mimetics. The following drugs were used: 5-methoxy-N-N-dimethyl-tryptamine and 8-hydroxy-2-(di-n-propylamino)tetralin hydrobromide, serotonin-agonists, fluoxetine and citalopram, inhibitors of serotonin uptake. All these serotonin-mimetics inhibit mouse killing behavior without apparent secondary effects. When these compounds were tested on killer rats, a stronger antimuricidal effect was observed in rats having altered serotonin neurotransmission. These results support a role for the serotoninergic supersensitivity in a model of aggressive behavior.

Agonist Inhibitor of 5-HT uptake Mouse killing behavior Raphe lesions Serotonin Serotonin-mimetics Supersensitivity

A reduced activity of central serotoninergic neurotransmission appears to be one of the critical factors underlying aggressive behavior [18, 23, 25, 33, 39]. Several reports have shown that defensive aggression induced by shock is enhanced by depletion of brain serotonin (5-HT) following 5,6- or 5,7-dihydroxytryptamine administration [7,15] or by a tryptophan-free diet [14]. Similarly, shock induced aggression has been facilitated after administration of a serotoninergic antagonist. Several authors have used mouse killing behavior (MKB) as a model of experimental aggression [17, 23, 24, 33, 34, 38, 39]. However, it has also been suggested that muricidal activity is a form of predatory behavior [33]. It was pointed out that food deprivation may facilitate MKB [1]. Nevertheless, this behavior is not induced by hunger [12] and hunger by itself is not a determinant for muricidal activity [16]. Rats with muricide reactions attack mice principally for the sake of killing [19] and such a behavior may be considered as a form of irritative aggression [2]. Several lines of evidence are in favour of an involvement of central serotoninergic mechanism in MKB [33,39]. Thus, selective 5-HT depletions following electrolytic or chemical lesions of the dorsal or median raphe nucleus—the region known to project forebrain serotoninergic innervation [3]—have been shown to induce MKB [4, 5, 10]. Similarly, several reports have described muricidal behavior following administration of an inhibitor of tryptophan hydroxylase, p-chlorophenylalanine (PCPA) which reduces brain 5-HT levels [6,9]. A reduction of 5-HT turnover in the raphe region in killer rats as compared to non-killer rats has also been reported [17, 23–25].

It is well founded that isolation induces aggressive behavior in DBA mice but not in C57. The latter shows a higher turnover of 5-HT [18]. Moreover, isolation induces a decrease of 5-HT turnover in both strains [18].

Thus, substantial evidence supports the involvement of a lack of inhibitory serotoninergic neurotransmission in MKB.

In view of these observations, a clear antimuricidal effect has been described following injection of 5-HT precursors [9], of 5-HT neuronal uptake inhibitors [6, 22, 28] or of 5-HT agonists [28].

Stimulation of central serotoninergic receptors induces stereotyped motor responses which include signs such as hindlimb abduction, forepaw treading and head weaving [13,36]. This pattern of behaviors has been used by several

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authors to investigate lesion-induced supersensitivity in serotoninergic projections [30,31].

In this paper, in order to explore whether or not changes in the sensitivity of 5-HT receptors may modulate MKB, we have investigated the effects of specific 5-HT agonists or of specific inhibitors of 5-HT uptake. The study reported has been carried out in spontaneous killer rats and in non-killer rats which became killer after alterations of the central serotoninergic system. It is observed here that the administration of PCPA or electrolytic lesions of dorsal and median raphe nucleus, were followed by potentiation of the effect of 5-HT mimetics on MKB. Data strongly suggest an involvement of 5-HT receptor supersensitivity.

METHOD

Animals

Male adult (2–3 months old) Wistar rats (250–350 g) from our colony were used in these experiments. They were maintained on a 12 hr light-dark cycle period (light on at 7 hr) with food and water freely available. They were housed individually in plastic opaque cages (21×40×15 cm) during 1 month.

Mouse Killing Behavior

After one month of social isolation, rats were tested for muricidal activity at 10 a.m. To determine MKB of animals, a mouse was placed in the cage only for 5 min and only those rats which killed mice consistently were used and classified as spontaneous killer rats after social isolation (K rats). K rats usually killed the mouse by breaking its neck with a latency time of 3±1 min. When the mouse was killed, it was removed from the cage. Animals which did not kill mice in a 30 min test were considered spontaneous non-killer rats after social isolation (NK) and submitted to PCPA treatment or were lesioned in the dorsal and median nucleus of the raphe. All rats were kept isolated throughout the experiment.

PCPA Treatment

Isolated NK rats were submitted to 150 mg/kg intraperitoneal (IP) of PCPA daily during two days. Twenty-four hours after the last injection they were again tested for muricidal activity. Those rats which kill the mice in less than 5 min were considered as parachlorophenylalanine killer rats (PCPA-K). This muricidal activity was confirmed during 5 consecutive days with one muricidal test each day. Ten days after the last injection, PCPA-K animals were tested for MKB again and injected with the different drugs at the lowest doses affording significant inhibition of MKB in K rats. PCPA-K rats were never injected two times in the same experimental group. In some cases PCPA-K rats were treated with a drug and 10 days after injected with saline for another experimental group. PCPA-K rats were also analysed for the "5-HT behavioral syndrome" following 5-HT agonist treatment.

Midbrain Raphe Lesions

Isolated NK rats were anesthetized with 40 mg/kg IP sodium pentobarbital and electrolytic lesions were produced in the dorsal and median raphe. Lesions were carried out by passing a 2 mA cathodal current for 20 seconds through an electrode which had been lowered according to the following stereotaxic coordinates (with lambda as reference): AP=−0.3, ML=0 and 6.5 mm deep for the dorsal and 8 mm

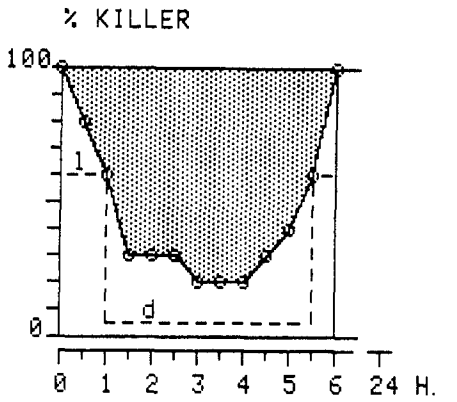

FIG. 1. Parameters of mouse killing inhibition. l: Latency time to reach 40% of inhibition of MKB. d: Duration of inhibition in at least 40% of muricidal rats tested. Efficacy (%) is the ratio of the filled area to the total area, areas are determined by weighing or by numerical integration.

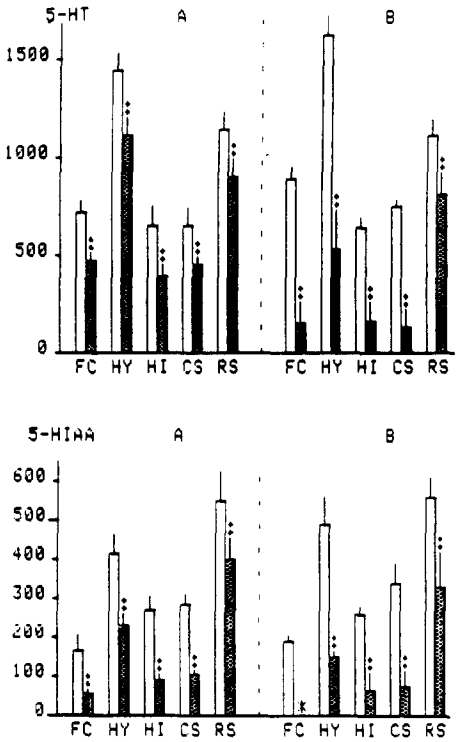

FIG. 2. Effect of lesioning the central serotoninergic system by a PCPA treatment (A) or by electrolytic lesion of the dorsal and median raphe nucleus (B) on the 5-HT and 5-HIAA contents of some rat brain structures. Rats were sacrificed 10 days after the last injection of PCPA or 50 days after midbrain raphe lesion. Values are expressed in ng/g wet weight. Mean values ±S.D. (N=6). Control rats: vehicle treated rats (/PCPA) or sham operated (/raphectomized). *Not detectable. ‡$p<0.001$ with respect to the control group (Student t-test). Structures: frontal cortex (FC), hypothalamus (Hy), hippocampus (HI), corpus striatum (CS), raphe slice (RS).

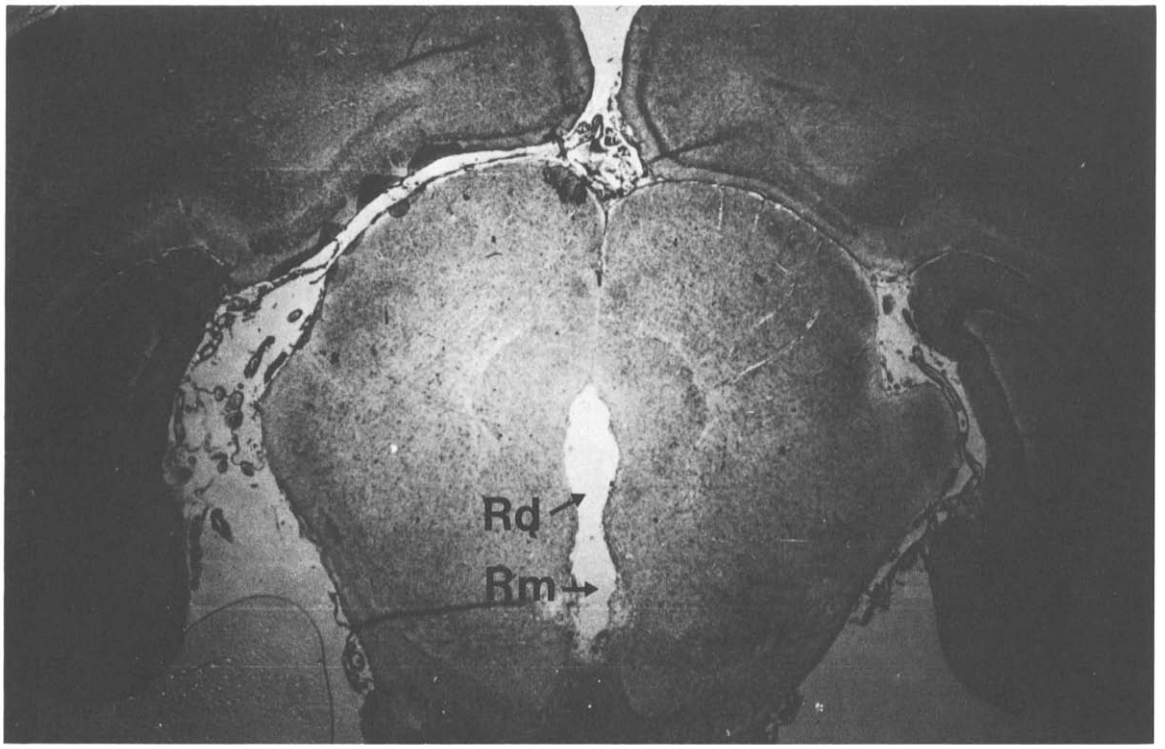

FIG. 3. Histological control of electrolytical lesions of raphe. Rd: raphe dorsalis; Rm: raphe medialis.

deep for the median raphe. Sham operated control animals were treated in the same way except that no current was passed through the electrode. Lesioned rats were individually housed for a recovery period of three weeks without any manipulation. At the end of this period all the animals were tested for MKB as described before. Those rats which killed the mouse in less than 5 min were considered as killer rats after raphe lesions (Ra-K); they were treated with the different drugs at the lowest doses affording significant inhibition of MKB in K rats and tested as mentioned earlier. Animals were also observed for the 5-HT behavioral syndrome following 5-HT agonist treatment.

5-HT Mimetic Treatment Procedures

The 3 groups of killer rats received the drugs in a randomized order and were then tested for muricidal activity. In each experiment, for each drug and dose, treated rats and control rats of the same group were tested in parallel. Animals were never injected two times in the same experiment. In come cases killer rats were treated with a drug and 10 days later injected with saline for another experiment. All drug administrations were performed by intraperitoneal injections (IP). After drug injections, animals were tested for their muricidal activity every 30 min during 6 hr and after 24 hr. In the case of 5-HT agonists, muricidal tests were carried out 15 min after drug administration followed by tests 30 min, 1 hr, 2 hr, and 3 hr after injection. Previous experiments have shown that for a killer rat its degree of experience with mouse killing has no influence on MKB. All the drugs were dissolved in saline and injected in a volume of 2 ml/kg. In the case of 5-HT agonist treatment, we also tested if injected animals show the 5-HT behavioral syndrome, since it is well known that stimulation of 5-HT receptors produces a pattern of behavior consisting of body flat posture, tremor, body shakes, head weaving and forepaw treading [13,36].

Parameters of Inhibition of Mouse Killing Behavior

Muricidal inhibition was measured by the following parameters:

(a) Percentage of K rats which no longer kill at different times after drug administration.

(b) Latency of inhibition of MKB (latency), time necessary for MKB to decrease to 60% in the tested rats as determined from the experimental kinetic curve. Two groups of 10 rats, one of which exhibiting 100% of MKB, the second exhibiting 60% of MKB, are significantly different at the 0.05 level according to the Fisher exact probability test (Fig. 1).

(c) Maximal percentage of mouse killing inhibition (maximum) after drug administration (Fig. 1).

(d) Efficacy: the efficacy (%) was determined as the ratio of the area of the inhibition surface (surface above the kinetic curve for MKB) to the total area (Fig. 1). The area was determined in the time interval 0–3 hr for agonists or in the time interval 0–6 hr for inhibitors of 5-HT uptake (Fig. 1).

(e) Duration of inhibition of MKB in at least 40% of rats tested (duration) (Fig. 1).

Locomotor Activity

Locomotor activity after the saline administration and under drug conditions was tested in an actograph apparatus. Tests began at 10 a.m. Each rat was placed in a Plexiglas box (100×20 cm) with 7 pairs of infrared photo cells 2 cm above the solid floor to provide an automated measure of locomotor activity. In each experimental group different killer rats were used for saline and the different doses of drug tested, number of rats per group=6. In the case of 5-HT agonists,

TABLE 1

EFFECTS OF A 5-HT AGONIST, 8-OH DPAT, ON MKB AND LOCOMOTOR ACTIVITY

Rats	Dose (μmol/kg IP)	N	Maximum (%)	Latency (min)	Duration (min)	Efficacy (%)	Locomotor Activity (Counts per 15 min) 0–15 min	15–30 min
K	0	10	0	—	—	0	121 ± 37	68 ± 21
	0.08	10	30	—	—	10	143 ± 52	61 ± 21
	0.16	10	40*	30	15	13	125 ± 43	55 ± 12
	0.32	10	80‡	15	45	25	123 ± 44	48 ± 10
PCPA-K	0	10	0	—	—	0	105 ± 19	62 ± 14
	0.08	9	77†§	10	30	36¶	98 ± 12	53 ± 14
	0.16	6	100‡§	15	45	27¶	108 ± 23	52 ± 15
Ra-K	0	9	0	—	—	0	135 ± 32	51 ± 21
	0.08	6	33	—	—	18§	119 ± 15	38 ± 20
	0.16	9	67†	15	30	23¶	118 ± 22	44 ± 14

Parameters of MKB are determined as described in the Method section from an experiment with N rats; latency and duration are given when the inhibition of MKB is significant.

Locomotor activity: N=6. Mean ± standard deviation.

Intra-group comparison with respect to vehicle treated rats of the same group: *$p<0.05$, †$p<0.005$, ‡$p<0.0005$.

Inter-group comparison with respect to drug treated K rats at the same dose: §$p<0.05$, ¶$p<0.005$.

Significant differences in efficacy are only shown for inter-group comparisons.

rats were treated and immediately placed in the actograph, activity was evaluated by the number of photobeam interruptions/15 min during 30 min. In the case of 5-HT uptake inhibitors rats were administered the drug and placed in the actograph. Here the activity was evaluated by the number of photobeam interruptions at each 30 min interval during 2 hr. The experimental rat groups investigated were: K rats, PCPA-K rats and Ra-K rats.

Biochemical Assay

5-HT and 5-hydroxyindolacetic acid (5-HIAA) were determined simultaneously utilizing a reverse phase chromatography procedure with an electrochemical detection according to Kempf *et al.* [18] with minor modifications. Frozen samples of the different brain regions were weighed and homogenized in 0.1 N $HClO_4$ containing 6 mM Na-metabisulfite and 1 mM EDTA. The homogenates were centrifuged at 10,000×g for 20 min at 4°C. Aliquots of the supernatant were transferred into the HPLC system with a Wisp automatic injector (Waters). The HPLC system consisted of a LC4 Bioanalytical System amperometric detector with a glassy carbon working electrode, a Bondapak phenyl column (10 μm particle size, 300×3.1 mm i.d.) and a pump (Waters).

The potential was set at 900 mV (vs. Ag-AgCl reference). The flow rate was 1.4 ml/minute and the sensitivity was set at 5 nA/V (1 volt full scale). The mobile phase consisted of 5% methanol in 0.1 M Na-phosphate buffer pH 3.5, 0.1 mM EDTA, 3,4-dihydroxyhydrocinnamic acid (Aldrich) was used as internal standard. Saline and PCPA treated rats were sacrificed for monoamine assay ten days after the last injection. Lesioned and sham operated rats were sacrificed fifty days following midbrain raphe lesions.

Drugs

5-Methoxy-N,N-dimethyl-tryptamine (5-MeODM) (Sigma Chemical Co., St. Louis, MO)—molecular weight: 218. (±)-8-Hydroxy-2-(di-n-propylamino)tetralin hydrobromide (8-OH DPAT) (Research Biochemicals Inc., Wayland, USA)—molecular weight: 328. Fluoxetine hydrochloride (Lilly Research Centre Limited, Surrey, England)—molecular weight: 309. Citalopram (Lundbeck & Co., Copenhagen, Denmark)—molecular weight: 405.

Histology

At the end of the experiments one group of 8 killer rats, with midbrain raphe lesions, were sacrificed and their brains were removed, subsequently fixed in 10% formalin and embedded in paraffin. Twenty-micron sections were stained with cresyl violet and the extent of the lesion damage for each rat was assessed by microscopic examination.

Statistics

(a) Mean values for 5-HT or 5-HIAA or locomotor activity are compared for the significance of their differences by the Fisher-Student's *t*-test.

(b) The significance of the inhibition of MKB is determined by the Fisher exact probability test. According to this test, when the experimental group is composed of 10 rats and when the 10 control rats present MKB, the inhibition of MKB is significant, at the 0.05 level, if 4 rats, at least, are inhibited in the treated group (inhibition of MKB=40%). The significance of the inter group inhibition of MKB is also analysed by the Fisher exact probability test.

(c) Efficacy: efficacies which are analogous to the ratio of observed population to expected population have been analysed for the significance of their differences by the χ^2 test.

(d) Latency and duration: these 2 parameters are determined by interpolation on the kinetic curve of MKB at the 40% level of inhibition, as such they cannot be analysed statistically. Since they are good indicators of the kinetic of the inhibition of MKB they are included as such in the data.

TABLE 2

EFFECTS OF A 5-HT AGONIST, 5-ME ODM, ON MKB AND LOCOMOTOR ACTIVITY

Rats	Dose (μmol/kg IP)	N	Maximum (%)	Latency (min)	Duration (min)	Efficacy (%)	Locomotor Activity (Counts per 15 min) 0–15 min	15–30 min
K	0	10	0	—	—	0	121 ± 37	68 ± 21
	2.3	10	30	—	—	13	110 ± 19	70 ± 15
	4.6	12	33	—	—	20	99 ± 17	58 ± 8
	6.9	10	50*	15	45	28	117 ± 21	59 ± 17
PCPA-K	0	10	0	—	—	0	105 ± 19	62 ± 14
	2.3	10	70†	15	30	22§	107 ± 25	57 ± 15
	4.6	8	100‡¶	20	40	26	73 ± 13†	45 ± 11*
Ra-K	0	10	0	—	—	0	135 ± 32	51 ± 21
	2.3	8	62*	15	60	26¶	120 ± 18	47 ± 15
	4.6	10	60*	15	90	30§	113 ± 25	52 ± 20

For legend see Table 1.

TABLE 3

EFFECTS OF AN INHIBITOR OF 5-HT UPTAKE, FLUOXETINE, ON MKB AND LOCOMOTOR ACTIVITY

Rats	Dose (μmol/kg IP)	N	Maximum (%)	Latency (min)	Duration (min)	Efficacy (%)	Locomotor Activity (Counts per 30 min) 30–60 min	60–90 min	90–120 min
K	0	10	0	—	—	0	122 ± 31	41 ± 7	29 ± 10
	14	11	55*	30	30	19	115 ± 27	54 ± 18	33 ± 13
	28	12	75‡	20	120	37	66 ± 17†	35 ± 11	15 ± 4†
PCPA-K	0	9	0	—	—	0	164 ± 31	47 ± 6	34 ± 4
	14	9	67†	15	120	47¶	167 ± 20	49 ± 9	40 ± 5
Ra-K	0	10	0	—	—	0	122 ± 34	41 ± 7	18 ± 11
	14	10	90‡§	15	105	36¶	115 ± 30	54 ± 20	33 ± 14

For legend see Table 1.

RESULTS

Effects of PCPA Treatment or Midbrain Raphe Lesions on Behavior

Ten percent of naive rats become K rats after 1 month of social isolation, when the non-killer rats are treated with PCPA (2×150 mg/kg) 40% become killer rats (PCPA-K); at the beginning of the muricidal tests there is no significant difference of body weight in the PCPA-K rats when compared either to the PCPA-treated non-killer rats or to control rats. When midbrain raphe lesions are carried out in non-killer rats, 50% of these rats become killer rats (Ra-K). At the beginning of the muricidal tests there is no significant difference of body weight in Ra-K rats when compared to the lesioned non-killer rats. Nevertheless, the body weight of lesioned rats is lower than that of control non-lesioned rats.

5-HT and 5-HIAA Contents in Several Brain Regions After PCPA Treatment or Midbrain Raphe Lesions

The electrolytic lesions of the dorsal and median raphe nucleus induced a clear depletion of 5-HT and 5-HIAA content in all the brain regions studied 50 days after the lesion (Fig. 2). Since these structures are innervated by serotoninergic pathways originating from the dorsal or median raphe nucleus, the results are the effects of the lesion of both nuclei. The histological controls showed that the lesions were localized in both raphe nuclei (Fig. 3).

A decrease of 5-HT and 5-HIAA levels was observed ten days after the last injection of PCPA (Fig. 2). It may be mentioned that 20 days after 300 mg/kg of PCPA no alterations either on 5-HT levels or in catecholamine content were observed [38].

Effect of 5-HT Agonists and 5-HT Uptake Inhibitors on MKB of Spontaneous K Rats

A clear dose-dependent inhibition of MKB was observed after both agonist treatment, 5-MeODM or 8-OH DPAT (Tables 1, 2), without decrease in locomotor activity. For both agonists the maximal percentage of mouse killing inhibition was observed 15 min after the administration of the

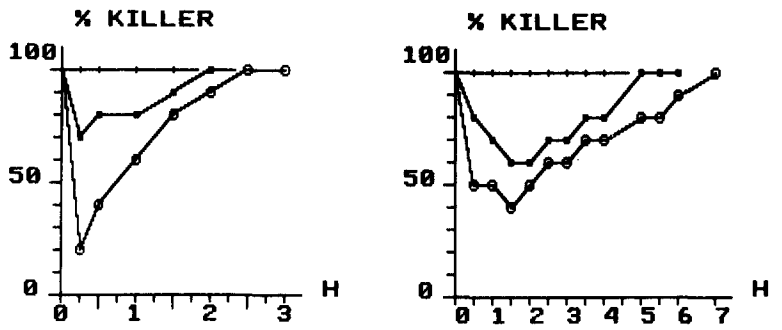

FIG. 4. Kinetic of inhibition of MKB after drug administration in K rats. Left: 8-OH DPAT (5-HT agonist). Control; 8-OH DPAT, 0.08 μmol/kg; 8-OH DPAT, 0.32 μmol/kg. Right: citalopram (inhibition of 5-HT capture). Control; citalopram, 2.5 μmol/kg; citalopram, 12.5 μmol/kg.

TABLE 4

EFFECTS OF AN INHIBITOR OF 5-HT UPTAKE, CITALOPRAM, ON MKB AND LOCOMOTOR ACTIVITY

Rats	Dose (μmol/kg IP)	N	Maximum (%)	Latency (min)	Duration (min)	Efficacy (%)	Locomotor Activity (Counts per 30 min) 30–60 min	60–90 min	90–120 min
K	0	10	0	—	—	0	122 ± 31	41 ± 7	29 ± 10
	2.5	10	40*	90	30	20	105 ± 20	48 ± 5	27 ± 3
	7.5	10	50*	60	60	23	110 ± 25	37 ± 6	22 ± 8
	12.5	10	60*	20	120	35	130 ± 25	44 ± 15	15 ± 5*
PCPA-K	0	10	0	—	—	0	164 ± 31	47 ± 6	34 ± 4
	2.5	10	80‡	30	60	32¶	168 ± 16	49 ± 7	32 ± 5
Ra-K	0	8	0	—	—	0	139 ± 56	56 ± 35	17 ± 8
	2.5	8	75†	30	180	42¶	130 ± 30	45 ± 9	26 ± 8*

For legend see Table 1.

drug. MKB returned to 100% 3 hr after the administration of the drug, an example of kinetic curves is given in Fig. 4. Confirming previous results [28] 8-OH DPAT, a putative ligand for 5-HT_{1a} receptor, induces a higher inhibition than 5-MeODM and at a much lower dosage. During the first 3–5 min after IP treatment, a slight body flat posture was observed with the highest dose of 5-MeODM.

Fluoxetine and citalopram, two compounds that block 5-HT uptake mechanism, blocked MKB in a dose-dependent way (Tables 3, 4) without decreasing locomotor activity but for the highest dose studied. For both inhibitors of 5-HT uptake the maximal percentage of mouse killing inhibition was observed 90 min after the administration of the drug, MKB returned to 100% 5–7 hr after the administration of the drug; an example of kinetic curves is given in Fig. 4. The 5-HT behavioral syndrome was not observed in our experimental conditions.

Effect of 5-HT Agonists and 5-HT Uptake Inhibitors on MKB of Killer Rats Obtained After PCPA Treatment

PCPA-K rats were treated by 5-HT uptake inhibitors or by serotonin agonists 10 days after PCPA administration. Similar to the spontaneous killer rats, both 5-HT agonists (8-OH DPAT and 5-MeODM) blocked MKB (Tables 1, 2). The antimuricidal effects observed following 8-OH DPAT or 5-MeODM administration were quite similar, however, the latter compound was injected at higher doses. During the first 5 min a slight body flat posture was induced by 4.6 μmole/kg of 5-MeODM. Fluoxetine and citalopram also inhibited MKB in PCPA-K rats (Tables 3, 4). For all the 5-HT mimetics the time at which the maximal percentage of mouse killing inhibition was obtained had not changed when compared to the control K rats. No decrease in locomotor activity was observed, but for the highest dose of 5-MeODM employed.

Effect of 5-HT Agonist and 5-HT Uptake Inhibitors on MKB of Killer Rats Obtained After Raphectomy

8-OH DPAT and 5-MeODM induced a dose-dependent antimuricidal effect on Ra-K rats (Tables 1, 2). At the highest dose 5-MeODM induced a slight body flat posture during the first 5 min; however, 15 min after 5-MeODM administration, while no body flat posture was observed, a significant decrease in MKB was seen. A significant lowering in MKB was observed at the lowest dose employed of 5-MeODM or of

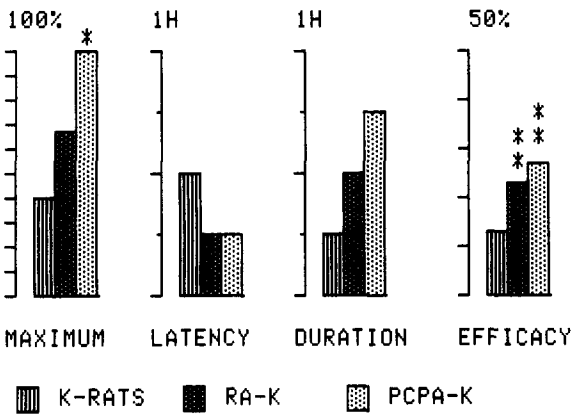

FIG. 5. Visualization of supersensitivity to a 5-HT agonist. Parameters of MKB (maximum, latency, duration, efficacy) in K rats and in rats in which serotoninergic neurotransmission has been altered (Ra-K and PCPA-K) after administration of 8-OH DPAT at the dose of 0.16 μmol/kg. Statistical analysis was performed on maximum and efficacy of Ra-K and PCPA-K as compared with K rats. $*p<0.05$; $**p<0.005$.

8-OH DPAT, whereas no sign of the 5-HT behavioral syndrome could be observed.

Fluoxetine and citalopram also blocked the MKB of Ra-K rats (Tables 3, 4). For all the 5-HT mimetics, the time at which maximal percentage of mouse killing inhibition did not change when compared to the control K rats. No reduction of locomotor activity was observed at the doses which were tested.

Comparison of the Effects of 5-HT Mimetic Drugs on MKB of K Rats, PCPA-K Rats, Ra-K Rats

In order to explore whether or not PCPA pretreatment or midbrain raphe lesions which produced serotoninergic neurotransmission alterations modified the response of killer rats to 5-HT mimetic drugs, the effects of these drugs have been compared at the same low dose(s) in the 3 groups of killer rats.

Inhibition of MKB has been followed by 4 parameters. Two among these could be statistically analysed for the significance of differences: maximum and efficacy. It appeared that the effects of the same dosage of 5-HT agonists or of inhibitors of 5-HT uptake were significantly stronger (higher maximum, higher efficacy) in PCPA-K rats and in Ra-K rats than in K rats, for at least one of these two parameters (Tables 1, 2, 3 and 4). Regarding the latency and duration, it could be observed that the latency was shorter and that the duration was longer in PCPA-K rats and in Ra-K rats than in K rats for the same dose of 5-HT agonists or inhibitors of 5-HT uptake (Tables 1, 2, 3 and 4). Figure 5 illustrates these effects for 8-OH DPAT as an example.

DISCUSSION

Both methods of alteration of 5-HT neurotransmission are known to decrease food intake and body weight gain [26]. In that case MKB induced by these treatments may be related to the altered appetitive states. Our data are in sharp contradiction to this hypothesis. Killer or non-killer rats in either PCPA treated or raphe lesioned rats do not show significant differences of body weight. Furthermore, if food deprivation may facilitate the induction of MKB [1], hunger by itself is not determinant for MKB [16].

Inhibition of MKB in K rats observed after serotonin-mimetic treatments confirmed the involvement of serotonin neurotransmission in muricidal behavior previously reported by numerous authors [7, 15, 23–25, 28, 33, 39]. 8-OH DPAT produced an inhibition of MKB without any sign of sedation or body flat posture. 5-MeODM at dosages above 4.6 μmol/kg produced a body flat posture in treated rats. However, this effect was observed 3 to 5 minutes after IP injection but not after 15 minutes when muricidal behavior was blocked. Moreover, at this time locomotor activity was normal and no sedative effect was observed. Thus a specific effect of serotonin agonists on MKB is strongly suggested. The known 5-HT uptake inhibitors, fluoxetine and citalopram, but for the highest doses used, reduced MKB without inducing secondary effects such as sedation or the 5-HT behavioral syndrome. As we have described before [28], 8-OH DPAT, an agonist postulated to be selective for 5-HT_{1a} receptors [29], induced at much lower dosage than 5-MeODM a higher decrease of MKB. Since global 5-HT affinity constants are very similar for both 5-HT agonists, it seems likely that the antimuricidal effect correlates with the activation of 5-HT_{1a} receptors. It is noticeable that both agonists show the same kinetic profile of inhibition of MKB. The maximal inhibition of MKB is observed 15 min after drug administration and the return to 100% MKB in 2–3 hr (Fig. 4). The 5-HT uptake inhibitors show also the same kinetic profile. In contrast with the agonists, the maximal inhibition is observed 90 min after drug administration and the return to 100% MKB in 5–7 hr. This explains why the MKB was not determined with the same time-schedule for agonists or for 5-HT uptake inhibitors. 5-HT is not the only neurotransmitter controlling MKB. We have previously shown that administration of GABA and serotonin mimetics inhibited MKB [27,28] and it is also known that α_1-adrenergic blockers inhibit MKB [35].

Determination of 5-HT content in different brain areas demonstrated a decrease of 5-HT and 5-HIAA levels 10 days after PCPA (2×150 mg/kg) administration in all the brain regions studied. This result is in agreement with those already published [20]. At this period we tested the first group of PCPA-K rats with 5-HT mimetic drugs. Muricidal behavior of PCPA-K rats was inhibited by 5-HT mimetic drugs. Moreover, a higher antimuricidal effect was observed in these rats, as compared to spontaneous K rats, submitted to similar dosages of 5-HT mimetic drugs. These data suggest a higher sensitivity to serotonin-mimetics in 5-HT depleted rats. In our experimental conditions, locomotor activity recordings do not show any sedative effect in PCPA treated rats but for the highest dose of 5-MeODM.

Lesions in the midbrain raphe induced MKB in previously non-killer rats. This confirmed observations already reported [4, 5, 10]. Biochemical studies show a great 5-HT depletion 50 days after midbrain raphe lesions in all the brain areas analysed, suggesting that both raphe nuclei were effectively lesioned. In our experiment both median and dorsal raphe nuclei were lesioned. This potentiates the percentage of Ra-K rats obtained [40]. 8-OH DPAT and 5-MeODM administration decreased muricidal activity of Ra-K rats. No sedative effect was observed following 5-HT agonist treatment in lesioned rats, suggesting that the observed inhibition of MKB was specific of the action of the serotonin-mimetics. In Ra-K rats again, the effect of serotonin mimetic drugs on MKB was stronger than in spontaneous killer rats. Reactivity is increased in PCPA treated or in midbrain raphe

lesioned rats [21,32]. Furthermore, the topology of bites in MKB is different in PCPA-K and Ra-K as compared to K rats [41]. Since inhibition of MKB in 5-HT depleted rats has been compared to that of K rats, the question arises whether this comparison is appropriate or not. In our opinion the comparison is appropriate: controls for the locomotor activity are killer rats of the same group, in each group the killer rats have been selected with the same criterion (a killer rat is a rat which kills consistently in less than 5 min). We are comforted in our opinion by the fact that the maximal inhibition of MKB after drug administration, agonists or uptake inhibitors, takes place at the same time for PCPA-K or Ra-K rats as for K rats. Thus, in two types of MKB induced either by PCPA treatment or raphe lesions, behavioral studies quite convincingly support the phenomenon of serotoninergic supersensitivity [30,31].

Electrophysiological experiments have clearly demonstrated that 5-HT denervation induced an enhanced responsiveness to microiontophoretically-applied 5-HT [39]. However, 5-HT binding studies after degeneration of serotoninergic pathways following either electrolytic lesion or 5,6- or 5,7-dihydroxytryptamine administration have led to some contradictory results. These discrepancies may be due to experimental conditions or to regional differences in *in vivo* modulation of 5-HT receptors [11]. Thus, the data of binding experiments do not lead to definitive conclusions. A higher inhibition of MKB in Ra-K rats, as compared to spontaneous K rats after 5-HT agonists treatment (8-OH DPAT or 5-MeODM) or inhibitors of 5-HT uptake (fluoxetine, citalopram), may reflect a behavioral expression of serotonin supersensitivity. Similar evidence for behavioral expression of serotonin supersensitivity was reported by Trulson *et al.* [37]. Once central serotoninergic nerve terminals were impaired by 5,7-dihydroxytryptamine, rats exhibited a supersensitivity in response to administration of serotonin precursors (L-5-hydroxytryptophan) or of agonists in a complex behavioral syndrome consisting of tremor, rigidity, hind limb abduction, Straub tail, lateral head weaving and reciprocal forepaw treading.

Another model for studying serotoninergic supersensitivity in brain was described by Fleischer *et al.* [8] after chronic treatment with PCPA. PCPA treatment resulted in enhanced supersensitivity to DL-5-hydroxytryptophan induced inhibition of response rates in rats working at a variable interval of 1 min schedule of milk reinforcement.

The serotoninergic supersensitivity we describe here in a type of aggressive behavior, seems to us an interesting model. This model involves supersensitivity in a behavioral model which can occur naturally and may explain differences in the capacities of animals to repress their aggressive pulsion.

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