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Pre- or Postsynaptic Activity of 5-HT_{1A} Compounds in Mice Depends on the Anxiety Paradigm

C. LÓPEZ-RUBALCAVA

Sección de Terapéutica Experimental, Departamento de Farmacología y Toxicología, CINVESTAV and División de Investigaciones en Neurociencias, Instituto Mexicano de Psiquiatría, Ap. Postal 22026, México 14000 D.F., México

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LÓPEZ-RUBALCAVA, C. *Pre- or postsynaptic activity of 5-HT_{1A} compounds depends on the anxiety paradigm.* PHARMACOL BIOCHEM BEHAV **54**(4) 677–686, 1996.—The purpose of the present study was to compare the contribution of pre and postsynaptic 5-HT_{1A} receptors to the anxiolytic effects of serotonergic$_{1A}$ compounds in two animal models of anxiety. To this aim, the 5-HT_{1A} ligands buspirone, ipsapirone, indorenate, and 8-OH-DPAT were tested in the burying behavior test and the avoidance exploratory behavior paradigm in control, pCPA-treated, and 5,7-DHT–lesioned mice. p-CPA and 5,7-DHT treatments did not modify the burying behavior per se, while 5-HT_{1A} agonists produced a significant reduction in this behavior in both p-CPA– and 5,7-DHT–lesioned animals. In the exploratory behavior paradigm, p-CPA per se but not 5,7-DHT increased the black/white transitions, interpreted as an antianxiety action. The ICV injection of 5,7-DHT blocked such effect of the 5-HT_{1A} compounds in the avoidance exploratory behavior test. Data suggest that the effect of 5-HT_{1A} compounds in the burying behavior test is mediated via the stimulation of postsynaptic receptors, while in the avoidance exploratory behavior paradigm these compounds act through the stimulation of the presynaptic site. Discussion is based on the differences between the animal models of anxiety.

5-HT_{1A} agonists p-CPA 5,7-DHT Pre- and postsynaptic receptors Mice Behavioral models of anxiety

SEVERAL lines of evidence support the idea that 5-HT_{1A} receptor ligands provide a novel class of anxiolytics [cf. (3,5,8,10,19)]. However, the exact mechanism through which these compounds mediate their anxiolytic activity remains unclear. It has been reported that 5-HT_{1A} receptors are located both, presynaptically (somatodendritic autoreceptors) on the 5-HT cell bodies in the dorsal and median raphe nuclei of the brain stem and postsynaptically, predominantly in limbic structures such as the hippocampus and the septum (33,47,48). Behavioral and metabolic effects of 5-HT_{1A} agonists appear to depend on the stimulation of either pre- or postsynaptic 5-HT_{1A} receptors (25,26,46). As to the antianxiety action of 5-HT_{1A} ligands, it is uncertain whether these compounds act through the stimulation of pre- or postsynaptic receptors. Reported data can be found in both senses. For example, some authors report that 5-HT_{1A} ligands mediate their anxiolytic activity through the stimulation of postsynaptic receptors (11,15,41) while others propose that these compounds act via presynaptic sites (12,34,37,38). The reasons for these controversies can be attributed to the different behavioral paradigms or species used in each study. Therefore, the purpose of the present work was to analyze whether the anxiolytic action of the 5-HT_{1A} agonists ipsapirone, buspirone, indorenate, and 8-OH-DPAT is mediated through the stimulation of pre- or postsynaptic 5-HT_{1A} receptors in mice, and to analyze if there are differences depending on the anxiety paradigm employed. To achieve this purpose, the anxiolytic effect of each of the 5-HT_{1A} compounds was compared between three groups: one group of animals depleted of serotonin by the administration of p-chlorophenylalanine (p-CPA), another group lesioned with the serotonergic neurotoxin 5,7-dihydroxytryptamine (5,7-DHT), and a control group (serotonin-intact animals). The experimental anxiety levels were established by means of the burying behavior test (35,45) and the avoidance exploratory behavior paradigm (6,7). To detect unspecific motor alterations, a general ambulatory test was carried out.

METHOD

Animals

Adult male Swiss Webster mice (20–30 g body weight) were used in this study. Animals were housed, five per cage,

in a room under controlled and inverted 12 L:12 D schedule (lights on at 2200 h). All animals had all over the experiment, ad lib access to water and Purina rodent chow.

Anxiety Tests

Burying Behavior Paradigm. This test consisted in placing the animal in a cage measuring 27 × 16 × 23 cm. The cage contained an electrified prod (7 cm long) emerging from one of its walls 2 cm above a fine sawdust bedding material. Every time the animal touched the prod it received an electric shock of 0.3 mA from a constant current shocker (La Fayette Instruments Co. model 5806). Immediately after placing the animal in the cage, its behavior was observed along 10 min. Once the animal received the first shock, it displayed the burying behavior characterized by moving a pile of bedding material with rapid alternating movements of its forepaws oriented to cover the aversive stimulus (electrified prod). The parameters registered in this anxiety test were: (a) the number of shocks previous to the display of the burying behavior; (b) the cumulative burying behavior, cumulative time (in seconds) that the animal spends burying the prod. A reduction in this behavior represents a decrease in anxiety, while the number of shocks are related to algesic actions (35,45).

Avoidance Exploratory Behavior Paradigm. This test was previously described by Crawley and Goodwin in 1980 and consists of a propylene test chamber measuring 44 × 21 × 21 cm darkened over one-third of its surface with black spray paint. An opening of 13 × 5 cm separated the dark third from the bright two-thirds of the cage. Fluorescent light above the cage illuminated the chamber's bright area. At the beginning of the test the animal was placed in the bright side of the cage and the number of transitions from one compartment to the other were recorded over a 10-min period. According to Crawley and Goodwin (6), an increase in the number of transitions from one side to the other is considered as an anxiolytic effect.

Activity Test

The ambulatory behavior was recorded in a box measuring 43 × 36 × 19 cm that was placed over a sensitive plaque (48 × 40 cm) of an activity meter (Stoelting Co., Chicago, IL) connected to a counter (Stoelting, Co). After the pharmacological treatment, the animal was placed in the cage and the number of counts were recorded over a 10-min period. Between each test the cage was carefully cleaned. The data are expressed as counts/10 min.

Drugs

The drugs used in this study were: 8-hydroxy-2-(di-n-propylamino) tetralin (8-OH-DPAT, Research Biochemicals Inc., Natick, MA), ipsapirone (Miles Pharmaceutical Division, West Haven, CO), buspirone (Mead Johnson, México City, México), indorenate (Department of Pharmacology, CINVESTAV, México City, México), p-chlorophenylalanine (p-CPA, Sigma, St. Louis MO), 5,7-dihydroxytryptamine (5,7-DHT, Sigma), and desipramine (Sigma).

p-Chlorophenylalanine was suspended in methyl-cellulose (15%); 5-HT_{1A} agonists were dissolved in physiological saline and desipramine in distilled water. These drugs were injected IP in a volume of 5.0 ml/kg. The neurotoxin 5,7-DHT was dissolved in ascorbic acid (0.2%) and administered ICV.

Experiment 1—Dose–Response Effect of 5-HT_{1A} Compounds in Naive Mice (Serotonin-Intact Animals)

In the anxiety tests used in this study, all animals received IP either saline or one of the four 5-HT_{1A} agonists: 8-OH-DPAT (0.25, 0.5 mg/kg; −20 min), ipsapirone (2.5, 5.0 mg/kg; 30 min), buspirone (2.5, 5.0, 10.0 mg/kg; −30 min) or indorenate (5.0, 10.0 mg/kg; −90 min). Doses and latencies for each of the serotonergic anxiolytics were established according to previous data (13,14). Independent groups for each anxiety paradigm were used. The data were statistically analyzed using one-way ANOVA test followed by Dunnett's *t*-test (43).

Experiment 2—Action of 5-HT_{1A} Ligands in Saline and p-CPA–Pretreated Mice

p-Chlorophenylalanine (600 mg/kg) was injected during 3 consecutive days. On the fourth, animals received either saline or one of the 5-HT_{1A} agents: 8-OH-DPAT (0.5 mg/kg; −20 min), ipsapirone (5.0 mg/kg; −30 min), buspirone (5.0 mg/kg; −30 min) or indorenate (10.0 mg/kg; −90 min). In this case the control group was injected with methyl-cellulose for 3 consecutive days and on the fourth treated similarly as the experimental groups. Data were statistically analyzed using Kruskal–Wallis analysis of variance followed by the Mann–Whitney *U*-test (40).

Experiment 3—Effect of 5-HT_{1A} Compounds in Sham and 5,7-DHT–Lesioned Animals

In a third set of animals, the serotonergic neurotoxin 5,7-DHT (75 μg in 4 μl) or ascorbic acid (0.2% in 4 μl) was injected into the right lateral ventricle of ether-anesthetized mice following the method described by Haley and McCormick (23). Previous to the ICV injection of the neurotoxin, animals were pretreated with desipramine (5 mg/kg, IP, −30 min) to protect noradrenergic neurones. Seven days later, animals received either saline or one of the 5-HT_{1A} compounds: 8-OH-DPAT (0.5 mg/kg; −20 min), ipsapirone (5.0 mg/kg; −30 min), buspirone (5.0 mg/kg; −30 min), or indorenate (10.0 mg/kg; −90 min). Also, in this case the data were statistically analyzed using the Kruskal–Wallis analysis of variance followed by the Mann–Whitney *U*-test (40).

After the anxiety experiments, all animals were submitted to the activity test previously described.

Neurochemical Analysis

With an independent group of animals a neurochemical analysis was performed. The mice belonging to this group were sacrificed by decapitation, the brain was removed, placed in a cold plate, and the brain stem and frontal cortex dissected. The tissue was placed in a vial containing 1.0 ml of 0.1 M perchloric acid and 0.05 mM ascorbic acid, frozen in liquid nitrogen and stored at −70°C until analyzed, at the most 36 h after dissection. Biogenic amines were determined by HPLC with electrochemical detection (2,28). After thawing, vials were spiked with 100 ng of 3,4-dihydroxybenzylamine (DHBA) as internal standard. Thereafter tissue was homogenized and the suspension centrifuged at 37,013 × *g* for 20 min at 4°C. Aliquots of 0.1 ml of the supernatant were injected into a liquid chromatography system (Waters Assoc., Milford, MA) and coupled to an amperometric detector (Bioanalytical Systems, West LaFayette, IN). The system was equipped with a reverse-phase column biophase ODS of 5 μm particle size (BAS) eluted with a mixture of 925 ml of 0.15 M monochloroacetic acid buffer, pH 3.0, containing 0.86 mM of sodium octyl sulphate, 75 ml of acetonitrile, and 18 ml of tetrahydrofuran. The column was kept at room temperature. Detection was carried out using a glassy carbon working electrode maintained at + 800 mV against Ag/AgCl, and the resulting current

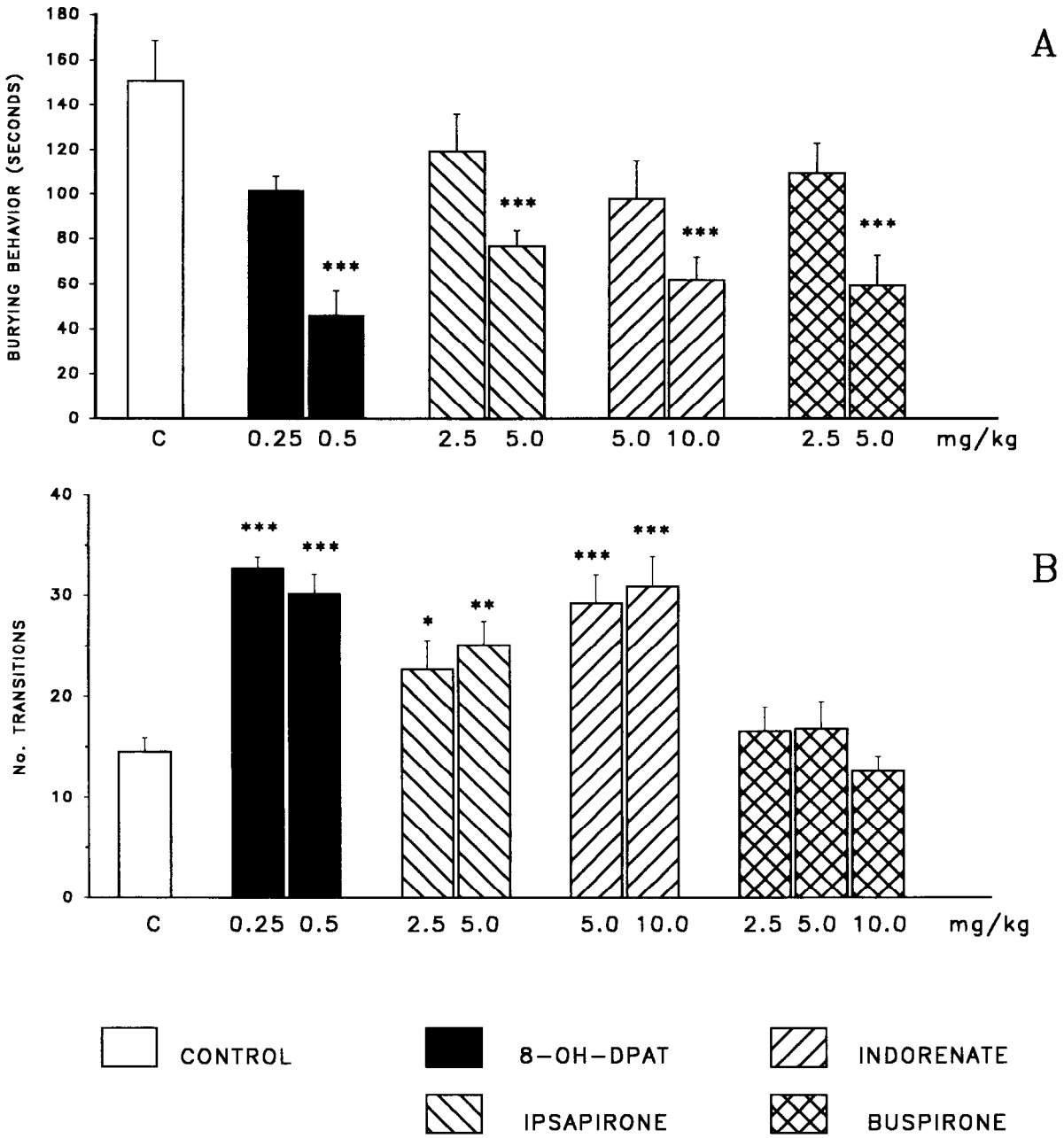

FIG. 1. Effect of the serotonergic anxiolytics 8-OH-DPAT, buspirone, ipsapirone, and indorenate in naive mice tested in the burying behavior paradigm (panel A) and exploratory behavior test (panel B). Figure shows mean ± SE ANOVA for burying behavior, $F(8) = 6.19$, $p < 0.05$; ANOVA for exploratory behavior, $F(8) = 10.14$, $p < 0.05$. Asterisks over columns represent statistical significant differences vs. the proper control group using the Dunnett's t-test, $^{*}p < 0.05$; $^{**}p < 0.02$; $^{***}p < 0.001$.

recorded. Retention times were 3.13, 8.30, and 11.93 min for noradrenaline (NA), 5-hydroxy-indolcacctic acid (5-HIAA) and serotonin (5-HT), respectively. The comparisons were performed between the p-CPA–treated group and its proper control and between sham operated and 5,7-DHT–lesioned mice. The data were statistically compared using the Student's t-test (40).

RESULTS

Figure 1 shows the effect of the 5-HT$_{1A}$ compounds in naive (serotonin-intact) mice. On panel A, it is clear that all 5-HT$_{1A}$ compounds produce a statistically significant decrease in burying behavior as compared with the saline treated group. On panel B, it can be seen that all 5-HT$_{1A}$ ligands, except for buspirone, induce a significant increase in the number of transitions between the black and white compartments as compared with the saline injected mice. After the results shown in this figure, the following doses were used: 8-OH-DPAT, 0.5 mg/kg; ipsapirone, 5.0 mg/kg; indorenate, 10.0 mg/kg, and buspirone 5.0 mg/kg.

Tables 1 and 2, summarize the neurochemical data obtained after the treatments with p-CPA and 5,7-DHT respectively.

TABLE 1

EFFECT OF *p*-CHLOROPHENYLALANINE (*p*-CPA) TREATMENT ON SEROTONIN (5-HT) AND 5-HYDROXY-INDOLEACETIC ACID (5-HIAA) LEVELS IN DIFFERENT BRAIN AREAS OF MALE MICE

		Cerebral Cortex	Brain Stem
5-HT	C	581.7 ± 46.2	708.2 ± 45.7
	T	285.1 ± 49.2†	339.7 ± 56.2†
5-HIIA	C	356.2 ± 18.3	463.8 ± 63.9
	T	261.2 ± 38.4*	159.9 ± 18.7†

Table shows mean ± SE of 5-HT and 5-HIIA content expressed as ng/g of tissue ($n = 10$ mice × group). Statistical comparisons were performed using the Student's *t*-test. *$p < 0.02$; †$p < 0.01$. C: vehicle treated group; T: *p*-CPA treated group.

TABLE 2

EFFECT OF 5,7-DIHYDROXITRYPTAMINE (5,7-DHT, ICV), ON SEROTONIN (5-HT), 5-HYDROXYINDOLEACETIC ACID (5-HIAA), AND NORADRENALINE (NA) LEVELS IN DIFFERENT BRAIN AREAS OF MALE MICE

		Cerebral Cortex	Brain Stem
5-HT	C	507.6 ± 58.3	795.5 ± 145.3
	T	267.8 ± 17.8†	393.2 ± 40.9*
5-HIIA	C	211.2 ± 26.6	437.7 ± 40.4
	T	140.3 ± 19.6†	353.5 ± 71.5 NS
NA	C	785.6 ± 43.7	737.9 ± 57.6
	T	660.4 ± 41.2 NS	645.8 ± 31.5 NS

Table shows mean ± SE of 5-HT, 5-HIIA, and NA content expressed as ng/g of tissue ($n = 10$ mice × group). Statistical comparisons were performed using the Student's *t*-test, NS, nonsignificant ($p > 0.05$), *$p < 0.02$; †$p < 0.01$. C: vehicle treated group; T: 5,7-DHT treated group.

A clear reduction in the levels of 5-HT and 5-HIIA in the brain stem and cerebral cortex was found after the administration of either p-CPA or 5,7-DHT; this last neurotoxin did not affect the levels of noradrenaline (Table 2).

Figure 2 shows the effect of the 5-HT_{1A} compounds in p-CPA treated (panel A) and 5,7-DHT lesioned (panel B) mice in the burying behavior test. Panel A compares the effect of the 5-HT_{1A} agonists ipsapirone (5.0 mg/kg), indorenate (10.0 mg/kg), 8-OH-DPAT (0.5 mg/kg), and buspirone (5.0 mg/kg) between methyl-cellulose and p-CPA–pretreated mice in the burying behavior paradigm. The administration of p-CPA per se did not modify the burying behavior when compared with the vehicle-treated animals (Mann–Whitney *U*-test, $p > 0.05$). In the case of the 5-HT_{1A} ligands, these compounds produced a significant reduction in the burying behavior of both control and p-CPA–treated animals. Finally, panel B shows the effect of the 5-HT_{1A} agonists in sham operated (ascorbic acid 0.2% ICV) and 5,7-DHT–lesioned animals. As in the case of p-CPA, the ICV administration of the neurotoxin per se did not modify the burying behavior when compared with the control group (Mann–Whitney *U*-test, $p > 0.05$); also, in this case all 5-HT_{1A} compounds produced a significant decrease in this parameter in both sham-operated and 5,7-DHT–injected animals.

Figure 3 shows the effect of the 5-HT_{1A} compounds in p-CPA treated (panel A) and 5,7-DHT lesioned (panel B) mice in the avoidance exploratory behavior test. Panel A shows the effect of 5-HT_{1A} agonists in control and p-CPA pretreated mice. Treatment with p-CPA per se produced a significant increase in the number of transitions from one compartment to the other when compared to the vehicle treated group (Mann–Whitney *U*-test, $p < 0.02$). In the same manner, 5-HT_{1A} agonists ipsapirone (5.0 mg/kg), indorenate (10.0 mg/kg) and 8-OH-DPAT (0.5 mg/kg) induced an increase in the number of transitions in the control group. However, in the p-CPA pretreated animals there was no statistical significant difference between the experimental groups and their respective control. In this case, buspirone was not evaluated because it did not produce any effect in this anxiety test (Fig. 1, panel B). Finally, on panel B, the action of the 5-HT_{1A} ligands in sham operated and 5,7-DHT lesioned mice on this test is shown. In this experiment, 5,7-DHT per se had no action (Mann–Whitney *U*-test, $p > 0.05$), while 5-HT_{1A} compounds produced a clear increase in the number of transitions in sham operated animals that was not observed in 5,7-DHT lesioned mice.

Figure 4 summarizes the effect of 5-HT_{1A} compounds administered to naive (panel A), pCPA (panel B) and 5,7-DHT (panel C) mice on the median number of shocks received before displaying burying behavior. As it can be seen, none of the treatments affected this parameter.

The results of the drug effects on the ambulatory behavior are shown in Table 3. The administration of p-CPA plus buspirone produced a significant reduction in general ambulatory behavior. Also, in sham-operated mice, 8-OH-DPAT and ipsapirone decreased general ambulatory behavior. In contrast, these two compounds increased this parameter in 5,7-DHT–lesioned mice. None of the other treatments affected ambulatory behavior.

DISCUSSION

In the present study, it was found that the systemic injection of the 5-HT_{1A} receptor compounds 8-OH-DPAT, ipsapirone, buspirone, and indorenate decreased burying behavior and increased the number of transitions between black and white compartments, both effects previously considered as anxiolytic actions (4,6,35,45). These anxiolytic-like effects of the serotonergic ligands have been reported in the presently used models (13,14,29,30) and in other models of anxiety (8,10,24), further supporting the suggestion that 5-HT_{1A} receptor ligands have anxiolytic properties (20,39,44). Because 5-HT_{1A} receptors are located both presynaptically in the raphe nucleus and postsynaptically in specific brain areas, it was an aim of the present study to investigate the relative contribution of these receptor populations to the antianxiety effects of 5-HT_{1A} receptor compounds.

In previous studies, it has been reported that the action of 5-HT_{1A} agonists can be blocked with p-CPA or 5,7-DHT when mediated by the activation of presynaptic receptors. For example, while using these treatments, it was found that the hyperphagic and hypothermic actions of 5-HT_{1A} compounds were mediated by presynaptic receptors (9,21). Present results suggest that in the burying behavior test, the antianxiety actions of the 5-HT_{1A} ligands are mediated via the stimulation of postsynaptic receptors. This conclusion arises from the fact that all 5-HT_{1A} agonists produced the same significant reduction in burying behavior in control, p-CPA–pretreated and 5,7-DHT–lesioned mice. These results are further sustained

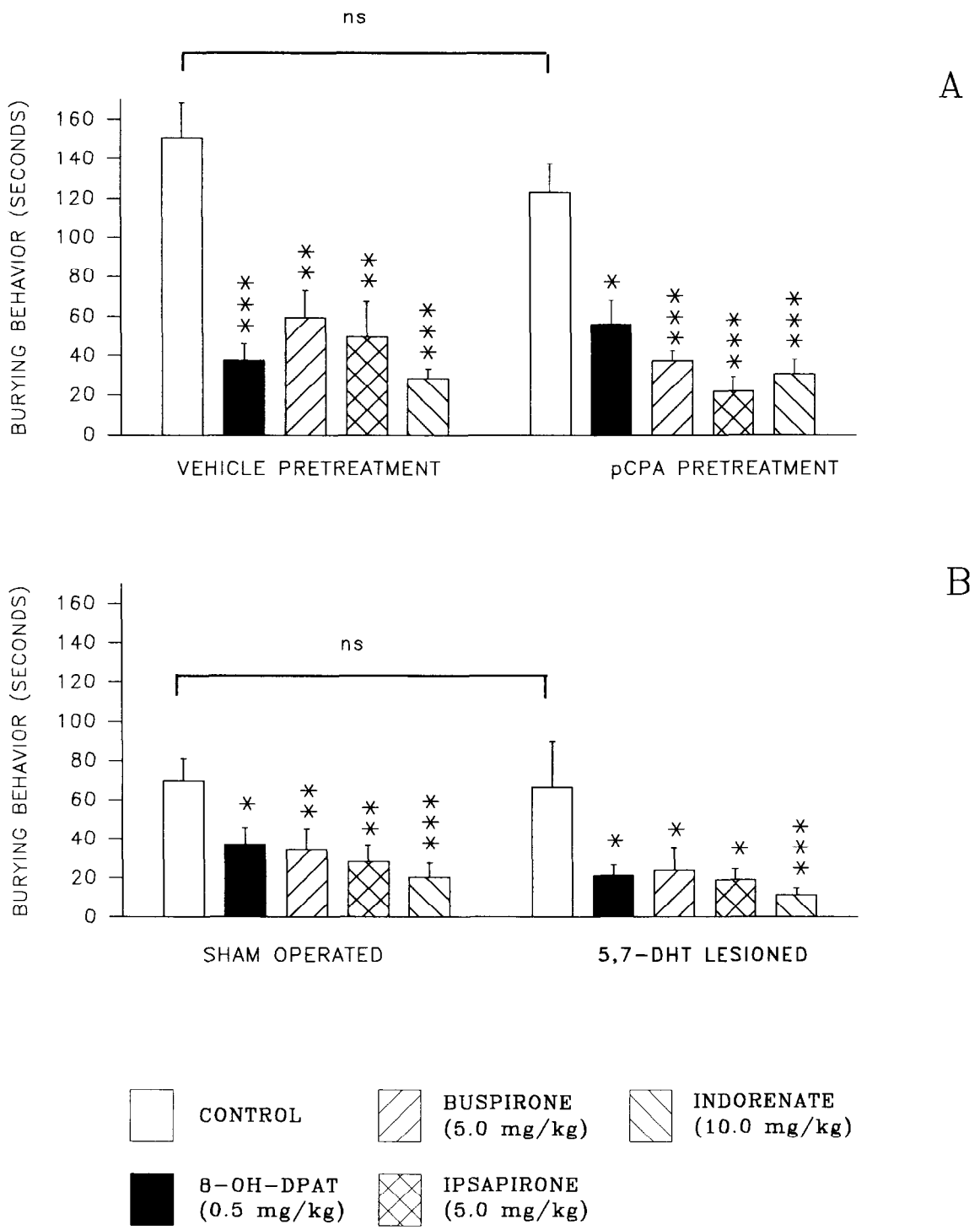

FIG. 2. Effect of the serotonergic anxiolytics 8-OH-DPAT, buspirone, ipsapirone, and indorenate in p-CPA pretreated (panel A) and 5,7-DHT lesioned (panel B) mice tested in the burying behavior paradigm. Figure shows mean ± SE time (s) of burying behaviour. Kruskal–Wallis analysis of variance for vehicle-pretreated group, $H(4) = 18.81$, $p < 0.001$; for p-CPA–pretreated group, $H(4) = 23.04$, $p < 0.001$; for sham-operated group, $H(4) = 12.18$, $p < 0.02$; for 5,7-DHT-lesioned group, $H(4) = 10.21$, $p < 0.05$. Mann–Whitney U-test between the 5-HT$_{1A}$-treated animals and their proper saline control group, ns = nonsignificant, $*p < 0.05$; $**p < 0.02$; $***p < 0.01$. The effects of p-CPA and 5,7-DHT per se are shown by a bracket.

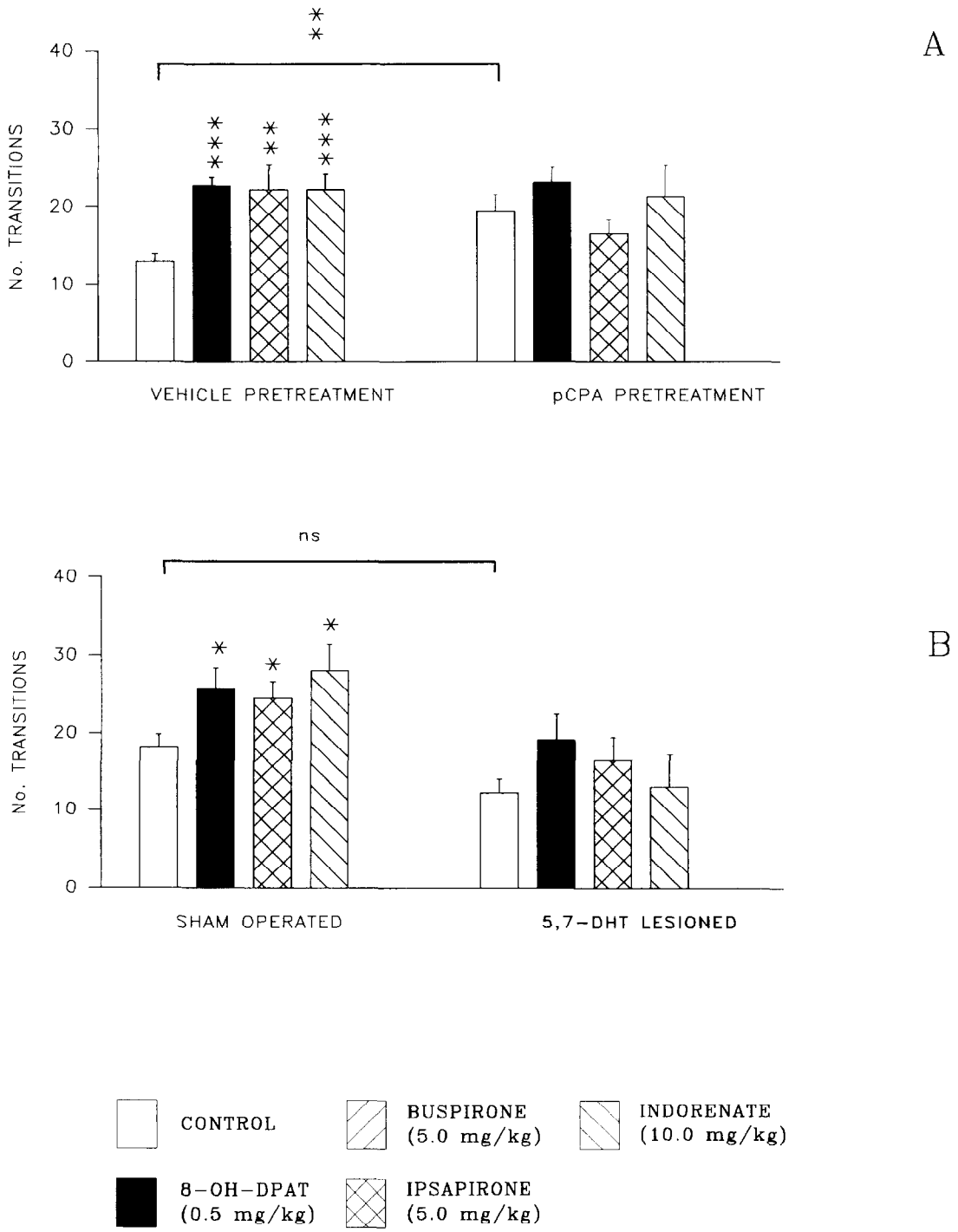

FIG. 3. Effect of the serotonergic anxiolytics 8-OH-DPAT, buspirone, ipsapirone, and indorenate in p-CPA pretreated (panel A) and 5,7-DHT lesioned (panel B) mice tested in the exploratory behavior test. Figure shows mean ± SE number of transitions. Kruskal–Wallis analysis of variance for vehicle-pretreated group, $H(3) = 15.52$, $p < 0.01$; for p-CPA–pretreated group, $H(3) = 3.74$, nonsignificant; for sham-operated group, $H(3) = 7.74$, $p < 0.05$; for 5,7-DHT–lesioned group, $H(3) = 3.31$, nonsignificant. Mann–Whitney U-test between the 5-HT_{1A}–treated animals and their proper saline control group, ns = nonsignificant, $*p < 0.05$; $**p < 0.02$; $***p < 0.01$. The effects of p-CPA and 5,7-DHT per se are shown by a bracket.

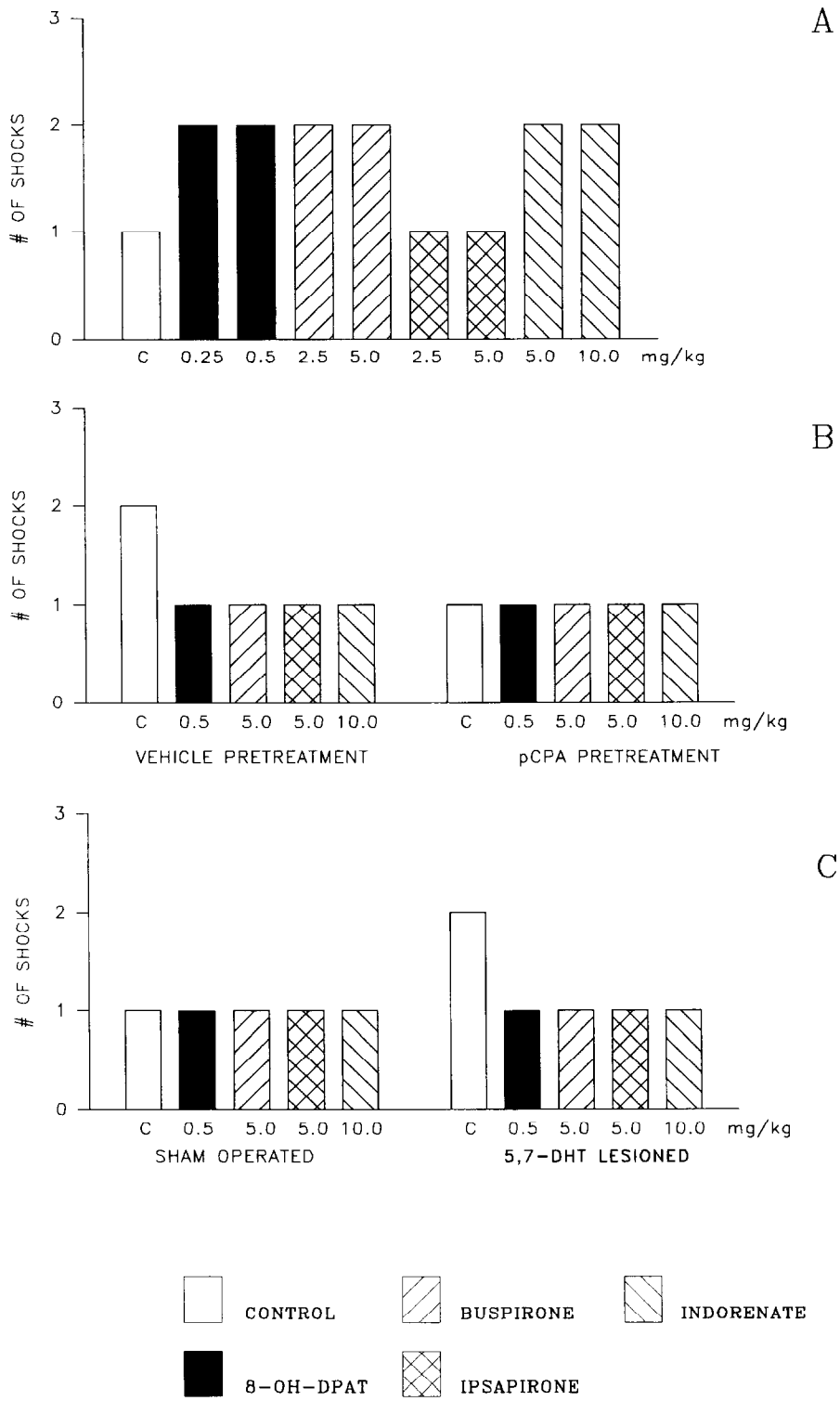

FIG. 4. Effect of the serotonergic anxiolytics 8-OH-DPAT, buspirone, ipsapirone, and indorenate in naive (panel A), p-CPA pretreated (panel B) and 5,7-DHT lesioned (panel C) mice tested in the burying behavior test. Figure shows median values of shocks received. ANOVA for naive mice, $F(8) = 1.04$, nonsignificant. Kruskal–Wallis analysis of variance for vehicle-pretreated group, $H(4) = 5.3$, nonsignificant; for p-CPA–pretreated group, $H(4) = 3.29$, nonsignificant; for sham-operated group, $H(4) = 2.11$, nonsignificant; for 5,7-DHT–lesioned group, $H(4) = 4.67$, nonsignificant.

TABLE 3

EFFECT OF 5-HT_{1A} AGONISTS IN MICE WITH DIFFERENT PRETREATMENTS IN THE MOTOR ACTIVITY TEST

Treatment (mg/kg)	Pretreatments				
	Naive	Methyl-cellulose	*p*-CPA	Sham-operated	5,7-DHT
	Number of Counts in 10 Minutes				
Saline	453 ± 63	400 ± 82	427 ± 77	460 ± 39	382 ± 38
8-OH-DPAT (0.5)	361 ± 57	361 ± 85	282 ± 59	341 ± 39*	421 ± 30*
Buspirone (5.0)	339 ± 63	339 ± 65	269 ± 45*	343 ± 45	257 ± 25
Ipsapirone (5.0)	421 ± 65	243 ± 39	278 ± 50	304 ± 26†	420 ± 71
Indorenate (10.0)	365 ± 62	462 ± 72	381 ± 52	482 ± 35	246 ± 34

Naive group: mice with no previous manipulation; Methyl-cellulose pretreated group: mice injected with methyl-cellulose (15%) for three consecutive days and on the fourth either saline or the 5-HT_{1A} agonists were administered; p-CPA–pretreated group: mice injected with p-CPA (600 mg/kg) for three consecutive days and on the fourth either saline or the 5-HT_{1A} agonists were administered; sham-operated group: seven days previous to the administration of either saline or the 5-HT_{1A} compounds, mice were injected with ascorbic acid (0.2 %, ICV) and desipramine (5 mg/kg, IP); 5,7-DHT lesioned group: mice that received 5,7-DHT (75 μg/4μl, ICV) and desipramine (5 mg/kg, IP), 7 days later animals received either saline or one of the 5-HT_{1A} compounds. Table shows mean ± SE number of counts per 10 min. Statistical comparisons were made between the 5-HT_{1A} treated groups and their respective saline control; also a comparison among all saline groups was made (nonsignificant). Mann–Whitney *U*-test, $*p < 0.05$; $\dagger p < 0.002$.

by previous studies in which ipsapirone, indorenate, and buspirone are reported to mediate their anxiolytic effect in the burying behavior test through the stimulation of postsynaptic receptors in rats (15). Although there are studies reporting species differences (particularly between mice and rats) in the anxiolytic action produced by 5-HT_{1A} agonists in this paradigm (14), it can be concluded that in both species the 5-HT_{1A} agonists ipsapirone, buspirone, and indorenate mediate their anxiolytic effect by the activation of postsynaptic receptors. However, it is important to mention that in relation to the antianxiety effect of 8-OH-DPAT there is a species difference in the site of action of this drug. In the present study, because the effect of 8-OH-DPAT was not blocked with the administration of p-CPA or 5,7-DHT, it can be assumed that this drug is acting through the stimulation of postsynaptic receptors; however, in a previous work we found that, in rats, the anxiolytic action of 8-OH-DPAT in the burying behavior test was mediated through the stimulation of presynaptic receptors (15). These results are in accordance with those reported by Bill et al. (1), who found that 8-OH-DPAT-induced hypothermia is mediated by presynaptic receptors in the mouse and by postsynaptic 5-HT_{1A} receptors in the rat. Further studies should be undertaken to analyze these species differences in the site of action of 8-OH-DPAT. Finally, it is also important to mention that in the burying behavior test the decrease in serotonergic transmission, produced either by p-CPA or by the ICV injection of 5,7-DHT, had no effect.

Opposed to the results obtained in the burying behavior model, in the avoidance exploratory behavior paradigm, p-CPA per se produced a significant increase in the number of transitions, an effect considered as a reduction in anxiety (6,7). Other authors have observed similar anxiolytic effects of p-CPA in different models of anxiety (16,18,36,42), suggesting that a reduction in serotonergic activity results in antianxiety effects. However, in the present study, the serotonergic lesion produced by 5,7-DHT has no anxiolytic action in this paradigm. The reasons underlying this discrepancy are unknown, although it has been reported that some behavioral effects of serotonin depletion depend on the method used (31).

As to the effect of the 5-HT_{1A} agonists in the avoidance exploratory behavior, it was found that these compounds produced a significant increase in the number of transitions in control animals. Because p-CPA has anxiolytic effects per se in this anxiety test, it is difficult to elucidate whether the antianxiety action observed with the 5-HT_{1A} ligands is mediated by themselves or by p-CPA. The lack of differences between the experimental and the p-CPA–pretreated control group might be reflecting a ceiling effect. Notwithstanding, when the serotonergic1A ligands were administered to 5,7-DHT–lesioned animals, these compounds lacked of an effect. These last results suggested that the 5-HT_{1A} drugs mediate their anxiolytic activity through the stimulation of presynaptic receptors in the avoidance exploratory test.

It is important to emphasize the differences observed in the anxiety animal models studied. In the avoidance exploratory behavior test p-CPA produces an anxiolytic effect per se. By contrast, in the burying behavior paradigm neither the serotonin depletion produced by 5,7-DHT nor with p-CPA resulted in a reduction of anxiety levels. Also, the 5-HT_{1A} compounds seem to act postsynaptically in the burying behavior test, while, in the avoidance exploratory paradigm, these drugs seem to act presynaptically in the mediation of their antianxiety actions. The main difference between these two anxiety paradigms is the nature of the aversive stimulus. In the burying behavior, the aversive stimulus is directly presented to the animal, while in the black and white transition test, the animal is confronted to a digress aversive stimulus (bright light). Also, these aversive stimuli produce different responses depending upon the anxiety model. Thus, while in the defensive burying anxiety test elicits the accomplishment of a behavior (burying behavior); in the avoidance exploratory paradigm, an inhibition of the behavior reveals anxiety (reduction in exploration). In accordance with this study, in the social interaction test (17), where bright light is used as an aversive stimulus that inhibits the expression of social behavior, it was dem-

onstrated that these 5-HT_{1A} compounds act presynaptically to produce anxiolytic effects (34). In support with this idea, Handley (24) reviewed the effects of serotonin-related drugs in anxiety models. She classified models according to the nature of the stimulus and of the response evoked, and found that the serotonergic drugs varied their actions depending on these parameters.

In a previous report, Broekkamp et al. (4) proposed that the anxiety animal models may reflect distinct types of anxiety disorders, which may be dissimilarly regulated. In clinical practice it has been reported that certain drugs are more effective in the treatment of some types of anxiety than others. For example, the antidepressants are more effective than benzodiazepines in agoraphobia and panic disorders (27,32). Beta blockers seem to be more useful for social phobia (22), while, serotonin-uptake inhibitors are the choiced drugs for obsessive-compulsive disorders (49). On these bases, it can be suggested that the animal models used in the present study may reflect different types of anxiety in which 5-HT might play different roles.

Finally, in the motor activity test, the combination of p-CPA plus buspirone produced a significant reduction in general ambulatory behavior. Hence, from these data, it is difficult to define whether the reduction in burying behavior observed with such combination is due to a specific anxiolytic effect or to an activity impairment. Also, in sham-operated mice, 8-OH-DPAT and ipsapirone decreased general ambulatory behavior. In contrast, these two compounds increased this parameter in 5,7-DHT–lesioned mice. These effects on ambulatory behavior seemed not to interfere with the action of these drugs in anxiety. This conclusion arises from the observation, that in the present study, when a drug-treatment decreases ambulatory behavior it also produces an increase in the number of transitions. Conversely, while drug treatment increases ambulatory behavior, in the avoidance exploratory test this procedure had no effect.

The present series of experiments demonstrate that 5-HT_{1A} compounds produce anxiolytic actions in mice after the stimulation of either pre- or postsynaptic receptors. The differences underlying the type of receptor population stimulated might rely primarily on the animal model of anxiety that concurrently depends on the nature of the aversive stimulus and/or the expression of the behavior characterized as anxiety.

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