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# Sex Behavior of Male and Female Wistar Rats Affected by the Serotonin Agonist 8-OH-DPAT

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HAENSEL, S. M., J. MOS, B. OLIVIER AND A. K. SLOB. *Sex behavior of male and female Wistar rats affected by the serotonin agonist 8-OH-DPAT.* PHARMACOL BIOCHEM BEHAV **40**(2) 221–228, 1991.—Four experiments were carried out to test the stimulatory effects of 8-OH-DPAT on various aspects of "masculine" sexual behavior of male and female rats and on the sexual attractivity of male rats. In Experiment 1 8-OH-DPAT (0.2 mg/kg) stimulated ejaculation frequency in middle-aged (approx. 15 months old) males, both sexually inactive and active subjects. There was a coinciding decrease in total number of mounts, intromissions, intromissions to first ejaculation and latency to first ejaculation. In Experiment 2 the effects of two doses (0.2 and 0.4 mg/kg) 8-OH-DPAT on the first ejaculation cycle were investigated. Especially, the higher dose made a high percentage (45–55%) of males to ejaculate "prematurely," i.e., at the first or second intromission. Latency to ejaculation decreased. With the higher dose, 25–35% of the males ejaculated extravaginally. In Experiment 3 8-OH-DPAT did not make males more attractive for an estrous female than saline-treated males, as judged by the time spent in their vicinity. However, estrous females received much more ejaculations from the tethered 8-OH-DPAT males, with the lowest latencies to first ejaculation, than from the saline-treated males. In Experiment 4 8-OH-DPAT stimulated mounting behavior in female rats only when they were long-term treated with testosterone. In that condition also shortest latencies to first mount were found with 8-OH-DPAT treatment.

$5\text{-HT}_{1A}$ 8-OH-DPAT Sexual behavior Masculine behavior Sexual attractivity Premature ejaculation Aphrodisiac Wistar rat

IT is now generally accepted that 8-hydroxy-2-(di-n-propylamino)tetralin (8-OH-DPAT) has stimulating properties on sexual behavior in male rats (2, 3, 21). At the time of the first publication, however, it was generally thought that serotonin was inhibitory (1). Thus the stimulatory effects of a serotonin agonist were quite surprising. The later developments in classifying $5\text{-HT}_1$ subtype receptor showed that 8-OH-DPAT is a powerful and probably full agonist at the $5\text{-HT}_{1A}$ site (22). Other $5\text{-HT}_{1A}$ agonists share the facilitatory effects on male sexual behavior (buspirone, ipsapirone, flesinoxan) thus suggesting the importance of this receptor for sexual behavior (13, 20, 23). In contrast, the $5\text{-HT}_{1B}$ site is thought to inhibit sexual behavior in rats (10). Detailed information on the effects of specific $5\text{-HT}_{1C}$ and $5\text{-HT}_{1D}$ agonists is presently not available. The results with 8-OH-DPAT and other $5\text{-HT}_{1A}$ agonists have stimulated further research into the neuropharmacological mechanisms underlying motivational and ejaculatory processes in sexual behavior (25).

The present experiments were carried out to replicate and elaborate some of the earlier reported findings, both in male and female rats (14,25). Also the possible stimulatory effects of 8-OH-DPAT on male sexual attractivity for a female partner were studied.

## GENERAL METHOD

### *Animals and Treatment*

Albino Wistar rats (HSD, Zeist, Holland) were used, except the male rats in Experiments 2 and 3, which were F1-hybrids of two inbred Wistar strains (R × U). The animals were housed two or three to a cage, of the same sex and treatment. Water and food were available ad lib. The day-night cycle was artificially maintained (dark 7:30 a.m.–5:30 p.m.) and temperature ranged from 22 to 24°C. Ovariectomies were carried out under light ether anesthesia via bilateral lumbar incisions. Stimulus females were brought in behavioral estrus by injecting 30 μg estradiol benzoate (EB) in 0.15 ml olive oil 48 or 24 h prior to testing, followed by 2.5 mg progesterone (P) in 0.1 ml oil 3 h before testing. A fresh solution of 8-OH-DPAT [(±) 8-OH-DPAT·HBr; Research Biochemicals Inc., Natick, MA] was made approximately 1 h before testing: 0.2 or 0.4 mg 8-OH-DPAT dissolved in 0.2 ml saline per kg body weight was administered by SC injection in the neck, 30 min prior to testing. Behavioral tests started about two hours after the onset of the dark cycle and the animals were allowed to adapt to the test cage for 15 minutes.

### *Behavioral Tests*

In pair-tests (Experiments 1, 2 and 4) behavioral testing was carried out in a semicircular arena, measuring 62 × 40 × 36 cm, with a wire mesh floor and a transparent front [e.g., (8)]. In the partner preference test (Experiment 3) a test box made of grey perspex with transparent front was used (27). It consisted of three compartments (60 × 30 × 40 cm each) with small openings

(13 × 12 cm) in both partitions near the front window. Each opening could be closed by a sliding door. Stimulus animals were put into the lateral compartments, one male treated with 8-OH-DPAT and one control male treated with saline. These males were either placed behind a wire mesh separation halfway the lateral compartment (tests 1–4) or given a leather harness which was attached with a stainless steel wire to the rear of the compartment to limit the action radius (tests 5–8). They were adapted to the test cage and tethering device twice for 15 minutes in the week before testing. The test room was dimly illuminated with indirect white light (60 W).

*Statistical Analysis*

Generally, data were subjected to one- or two-way analyses of variance (ANOVA) for repeated measures (24). Significant interactions were analysed with the simple main effects (SME) method (15). If the overall test was statistically significant, the Least Significant Difference (LSD) Test was used to make pairwise comparisons among means (15). Friedman's two-way ANOVA was used for nonparametric analysis (26). The 0.05 level of probability was adopted as the level of statistical significance.

## EXPERIMENT 1: DOES 8-OH-DPAT STIMULATE SEXUAL BEHAVIOR IN MIDDLE-AGED MALE RATS?

It has been shown that 8-OH-DPAT increases ejaculatory performance in male rats (2, 4, 21, 25). Such studies employed males that were approximately 6 months old, i.e., relatively young. It was thought to be of interest to investigate the sexually stimulatory properties of the drug in middle-aged male rats, particulary because such rats are assumed to have impaired reproductive capabilities and are generally assumed to be less sexually active (12, 16, 17).

### METHOD

Twelve middle-aged male rats (12–14 months old at the start of the experiment; body weights between 540 and 695 g) were tested for masculine sexual behavior. These animals were former breeding males but had not had heterosexual experience for at least four months prior to testing. Following the first 15-min pair-test two groups were formed on the basis of their sexual performance: an "active group" (n = 7), consisting of males that ejaculated at least once and an "inactive group" (n = 5) of males that did not ejaculate.

The animals were tested once a week with an estrous female for 7 consecutive weeks. When a stable level of sexual behavior was found (i.e., tests 3 and 4), treatment with 8-OH-DPAT or saline was started. In tests 5, 6 and 7 the animals were treated with saline (2 ml/kg), 8-OH-DPAT (0.2 mg/kg) and saline (2 ml/kg), respectively. Finally, test 8 (saline) was given 4 weeks following test 7.

### RESULTS

Ejaculation behavior is shown in Fig. 1. During 8-OH-DPAT treatment there was a clear stimulation of the mean number of ejaculations in both groups of males. Statistical analysis (two-way ANOVA on tests −1 to +5) revealed an effect of tests, $F(3,30) = 8.752$, $p < 0.001$; $LSD(5\%) = 0.61$, a borderline significant effect of groups, $F(1,10) = 4.177$, $p < 0.07$, and no significant interaction, $F(3,30) = 0.47$, n.s. From inspection of the results it is clear that during 8-OH-DPAT treatment all males of both groups ejaculated with high frequencies. During 8-OH-DPAT treatment males of both groups did not differ in ejaculation frequencies, which were in both groups significantly higher than during the tests before and after.

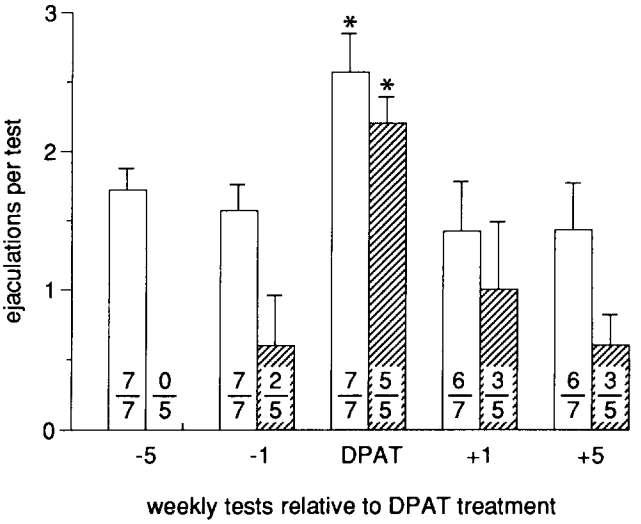


FIG. 1. Mean (±SEM) ejaculation frequency of initially active (n = 7; open bars) and inactive (n = 5; hatched bars) middle-aged male rats before and after 0.2 mg/kg 8-OH-DPAT treatment. Males were tested weekly for 7 consecutive weeks; the results of the 1st (−5), 5th through 7th (−1; DPAT; +1) and the 8th (+5) test (4 weeks following test 7) are depicted. Asterisks indicate a significant difference from tests before and after treatment (see also Table 1). Also indicated is the number of animals per test that ejaculated at least once.

Various other sexual behavior parameters are presented in Table 1. With regard to mounting behavior it was found that for both groups of males lowest frequencies were observed during tests with 8-OH-DPAT treatment [two-way ANOVA on tests −1 to +5, groups, $F(1,10) = 1.01$, n.s.; tests, $F(3,30) = 4.31$, $p < 0.02$; groups × tests interaction, $F(3,30) = 2.16$, n.s.; $LSD(5\%;\text{tests}) = 4.32$]. For intromission frequencies similar results were obtained, i.e., lowest frequencies with 8-OH-DPAT treatment [two-way ANOVA, groups $F(1,10) = 0.62$, n.s.; tests $F(3,30) = 2.10$, n.s.; groups × tests interaction $F(3,30) = 3.08$, $p = 0.04$]. It is clear that the number of intromissions to first ejaculation did not seem to differ between the two groups of males; lowest numbers were found during 8-OH-DPAT treatment (see also Experiment 2).

Two-way ANOVA of mount latencies (tests −1 through +5) did not reveal significant differences [groups, $F(1,10) = 3.38$, $p = 0.096$; tests, $F(3,30) = 0.89$, n.s.; groups × tests interaction, $F(3,30) = 1.33$, n.s.]. Latency to first ejaculation differed significantly over the tests [$F(3,30) = 9.46$, $p < 0.001$, $LSD(1\%) = 246.0$], but the groups of males did not differ, $F(1,10) = 2.65$, n.s., and there was also no significant interaction, $F(3,30) = 1.02$, n.s.

Thus there was a trend for an overall group difference for latency to first mount, while the males did not differ for first ejaculation latency. Both groups showed shortest latencies to ejaculation with 8-OH-DPAT treatment.

## EXPERIMENT 2: "PREMATURE EJACULATION" CAUSED BY 8-OH-DPAT

It has been noted that with a high dose of 8-OH-DPAT male rats can ejaculate already at the first intromission [e.g., (2)].

TABLE 1

SEXUAL BEHAVIOR PARAMETERS (MEANS ± SEM) IN SEXUALLY ACTIVE (A; n=7) AND INACTIVE (I; n=5) MIDDLE-AGED MALE WISTAR RATS

| Behavior | | Weekly Tests (n) −5 | (n) −1 | (n) 8-OH-DPAT | (n) +1 | (n) +5 | Statistics (two-way ANOVA) on last 4 tests; only significant F values presented (see also text) |
|---|---|---|---|---|---|---|---|
| Mounts | | | | | | | |
| (number) | A | (7) 12.1 ± 2.3 | (7) 15.0 ± 3.9 | (7) 7.3 ± 2.3 | (7) 13.7 ± 3.8 | (7) 10.4 ± 2.7 | Tests, $F(3,30)=4.31$, $p<0.02$, |
| | I | (2) 11.6 ± 6.4 | (5) 18.0 ± 2.2 | (5) 8.8 ± 1.9 | (5) 13.4 ± 2.6 | (5) 22.2 ± 4.7 | LSD(5%)=4.32 |
| Intromissions | | | | | | | |
| (number) | A | (7) 13.0 ± 1.1 | (7) 13.3 ± 1.4 | (7) 9.0 ± 1.0 | (7) 10.6 ± 1.7 | (7) 12.9 ± 1.0 | Groups × Tests, $F(3,30)=3.1$, |
| | I | (2) 2.4 ± 1.4 | (5) 10.8 ± 2.3 | (5) 9.2 ± 0.8 | (5) 13.2 ± 2.0 | (5) 7.6 ± 1.5 | $p=0.04$ |
| number to first ejaculation | A | 8.4 ± 1.0 | 10.1 ± 1.3 | 4.4 ± 0.9 | 7.3 ± 2.4 | 8.7 ± 0.5 | † |
| | I | — | 12.0 ± 0.7 | 3.9 ± 1.2 | 8.7 ± 1.0 | 9.7 ± 1.1 | |
| Ejaculations | | | | | | | |
| (number) | A | (7) 1.7 ± 0.2 | (7) 1.6 ± 0.2 | (7) 2.6 ± 0.3 | (6) 1.4 ± 0.4 | (6) 1.4 ± 0.4 | Tests, $F(3,30)=8.8$, $p<0.001$, |
| | I | (0) 0 | (2) 0.6 ± 0.4 | (5) 2.2 ± 0.2 | (3) 1.0 ± 0.5 | (3) 0.6 ± 0.2 | LSD(5%)=0.61 |
| | | | | | | | Groups, $F(1,10)=4.2$, $p<0.07$ |
| Latencies (s)* | | | | | | | |
| to first mount | A | 20.0 ± 5.5 | 7.3 ± 1.0 | 7.1 ± 1.1 | 8.1 ± 1.6 | 9.1 ± 1.4 | Groups, $F(1,10)=3.38$, $p=0.096$ |
| | I | 578 ± 281 | 27.2 ± 12.0 | 10.2 ± 1.9 | 27.8 ± 15.0 | 12.7 ± 2.4 | |
| to first ejaculation | A | 360 ± 37 | 393 ± 40 | 122 ± 62 | 511 ± 111 | 520 ± 109 | Tests, $F(3,30)=9.46$, $p<0.001$, |
| | I | 900 | 763 ± 113 | 217 ± 101 | 595 ± 156 | 673 ± 96 | LSD(5%)=246.0 |

(n) Number of Ss displaying the behavior.
*Nine hundred s for nonresponders.
†Responders only; data not suitable for ANOVA, see also Experiment 2.
Weekly pair-tests (15 min) with estrous female relative to DPAT treatment.

This phenomenon, which may be called "premature ejaculation" by analogy with what is described in the human [e.g., (19)], was investigated in more detail. Questions that were studied comprised firstly the quality of the ejaculate, whether or not it contained mobile spermatozoa, secondly where the semen was deposited, i.e., intra- or extravaginally and thirdly the details of the sexual behavior.

## METHOD

Twenty heterosexually naive male rats, approximately 6 months old, were pair-tested for sexual behavior. Tests lasted till one ejaculation had occurred. The numbers of mounts and intromissions prior to ejaculation were scored, and latencies to first mount (ML), first intromission (IL) and ejaculation (EL) were calculated. Immediately after ejaculation of the male the stimulus female was removed from the test cage and a vaginal smear was taken, which was subsequently studied microscopically. The test cage was carefully searched for the presence of a seminal plug; both animals were also thoroughly inspected for possible ejaculate coagulated to their furs.

Five consecutive weekly tests were carried out in which the animals received saline (2 ml/kg), a low dose 8-OH-DPAT (0.2 mg/kg), a higher dose 8-OH-DPAT (0.4 mg/kg), saline (2 ml/kg) and again the higher dose 8-OH-DPAT (0.4 mg/kg), respectively.

## RESULTS

Various sexual behavior parameters are presented in Table 2. The main findings are that latency to ejaculation is significantly shorter with DPAT treatment. Mean number of intromissions prior to ejaculation and latency to first intromission decreased significantly with 8-OH-DPAT treatment. Although lowest values were found with the higher dose, this was not significantly different from the lower dose 8-OH-DPAT. "Premature ejaculations" (i.e., ejaculations at first or second intromission) were never observed following saline injection (tests 1 and 4). With the low dose of 8-OH-DPAT one male ejaculated during the first and eight males during the second intromission. With the higher dose of 8-OH-DPAT (tests 3 and 5), 11 and 10 males, respectively, ejaculated during their first or second intromission.

With regard to the location of the semen it was found that this was usually the vagina. However, with the higher dose 8-OH-DPAT (tests 3 and 5) 7 and 5 animals, respectively, were found to ejaculate extravaginally. Surprisingly, this was also observed in one male in test 1 and one other male in test 4.

The quality of the ejaculate, as judged by microscopic inspection revealed no systematic differences between the different tests. Live spermatozoa were always found.

## EXPERIMENT 3: DOES 8-OH-DPAT RENDER MALES MORE SEXUALLY ATTRACTIVE TO FEMALES?

When given the choice in a 3-compartment box between 2 tethered stimulus males, one intact and one castrated, an estrous female rat prefers the vicinity of the castrated male (7,9). Females' preference for the castrated male was mainly due to the aversion to genital stimulation received during intromissions by the intact male. When intromissions were prevented through vaginal occlusion, intact males became by far the preferred part-

TABLE 2

SEXUAL BEHAVIOR PARAMETERS (MEANS ± SEM) IN MALE RATS (n = 20) TESTED TILL THE FIRST EJACULATION

| | Test 1 Saline | Test 2 8-OH-DPAT 0.2 mg/kg | Test 3 8-OH-DPAT 0.4 mg/kg | Test 4 Saline | Test 5 8-OH-DPAT 0.4 mg/kg | Statistics (one-way ANOVA) F(4,76)-value LSD (5%) value |
|---|---|---|---|---|---|---|
| Intromissions: | | | | | | |
| number | 12.7 ± 0.6 | 2.4 ± 0.6 | 1.6 ± 0.4 | 14.0 ± 1.1 | 1.8 ± 0.3 | $F=7.54$; $p<0.001$ |
| range | 8–18 | 0–14 | 0–5 | 6–26 | 0–4 | LSD = 1.45 |
| Latency to first intromission (s) | 76.1 ± 24.7 | 28.9 ± 10.4 | 6.8 ± 1.3 | 14.7 ± 2.2 | 12.2 ± 4.4 | $F=6.05$, $p<0.001$ LSD = 27.03 |
| Ejaculation: | | | | | | |
| latency (s) | 505.4 ± 52.0 | 122.9 ± 46.6 | 35.9 ± 13.2 | 289.1 ± 28.1 | 38.6 ± 8.9 | $F=37.03$, $p<0.0001$ |
| No. 1st intro-ejaculations* | 0 | 1 | 7 | 0 | 2 | LSD = 7.44 |
| No. 2nd intro-ejaculations* | 0 | 8 | 4 | 0 | 8 | |
| Ejaculate: | | | | | | |
| vaginal | 16 | 15 | 10 | 19 | 14 | |
| extravaginal | 1 | 1 | 7 | 1 | 5 | |
| combined | 2 | 1 | 2 | 0 | 0 | |
| not found | 1 | 3 | 1 | 0 | 1 | |

*Number of males ejaculating at first or at second intromission.
Five weekly tests, with DPAT or saline treatment prior to testing.

ners (7). 8-OH-DPAT treatment was reported to decrease mounting and intromission frequencies in male rats [(2, 3, 25); see also Experiments 1 and 2 of the present study]. Therefore, it was postulated that 8-OH-DPAT treatment could make males more attractive as a mating partner for an estrous female rat. This idea was tested in the following experiment.

## METHOD

Fifteen heterosexually naive female rats were tested for partner preference behavior in a 3-compartment box. They were ovariectomized at the age of 3 months and behavioral testing started one month later. They were brought in behavioral estrus by EB, 20 μg 48 and 24 h prior to testing, followed by P, 2.5 mg 3 h before the 15-min test. The stimulus males (n = 30) were treated with saline or 8-OH-DPAT (0.2 mg/kg b.wt.) 30 min prior to testing. At the start of the test the sliding doors were removed and the time spent in each compartment was recorded. To quantify partner preference, a preference score was calculated in which the time spent with the saline-treated male was subtracted from the time spent with the 8-OH-DPAT male. Thus a positive score indicates a preference for the 8-OH-DPAT male, while a negative score means that the control male is preferred. In tests 1–4 the males were kept behind wire mesh, whereas in tests 5–8 this wire mesh partition was removed and behavioral interaction was possible between the estrous female and the tethered males. During the latter 4 tests various sexual behavior data were recorded.

## RESULTS

### *Partner Preference Behavior*

The data of tests 1–8 were subjected to one-way ANOVA. There appeared to be no significant preference for either partner, $F(7,98)=1.28$, n.s.; see Fig. 2. The time spent in the empty middle compartment revealed interesting results. One-way ANOVA (tests 1–8) showed a significant effect, $F(7,98)=14.43$, $p<0.001$. Further LSD analysis [LSD(5%) = 69.9; LSD(1%) = 99.3] indicated that during tests 3 and 4 (no behavioral interaction with stimulus males possible) the estrous females spent most of the time in the lateral compartments which contained the males. In tests 7 and 8 the estrous females spent about half of the total time in the middle compartment although in the remaining time they were sexually very active with the tethered males (see later, Figs. 3 and 4).

### *Sexual Behavior With Tethered Males*

Various sexual behavioral parameters are presented in Figs. 3 and 4. Two-way ANOVA of ejaculation frequencies (Fig. 3, top) revealed a significant effect of tests, $F(3,42)=19.39$, $p<0.001$, of treatment of the stimulus males, $F(1,14)=19.27$, $p<0.001$, but no significant interaction, $F(3,42)=0.37$, n.s. Thus the females received a progressively increasing number of ejaculations over the 4 tests, while overall the 8-OH-DPAT treated males were "preferred" in this respect. Only during test 5 (the first test with behavioral interaction possible), there was a significant difference between the number of 8-OH-DPAT- and saline-treated males which ejaculated 9/15 vs. 2/15, respectively (Fisher exact probability test $p=5.17$, $df=1$, $p<0.02$).

The mean latencies to first ejaculation (Fig. 3, middle; with 900-s cut-off value for nonejaculators), subjected to Friedman's two-way ANOVA, showed a significant effect of tests, $\chi_r^2(3)=15.12$, $p<0.001$, and treatment, $\chi_r^2(1)=19.27$, $p<0.001$, but no significant interaction was apparent. Hence, over the 4 tests, females received the first ejaculation progressively sooner after the beginning of testing, while the shortest latencies occurred with the 8-OH-DPAT-treated males.

Mean latencies to first ejaculation of responders only (Fig. 3, bottom) were significantly different between 8-OH-DPAT- and saline-treated males in tests 6, $t(11)=2.31$, $p<0.05$, and 8, $t(24)=2.28$, $p<0.05$.

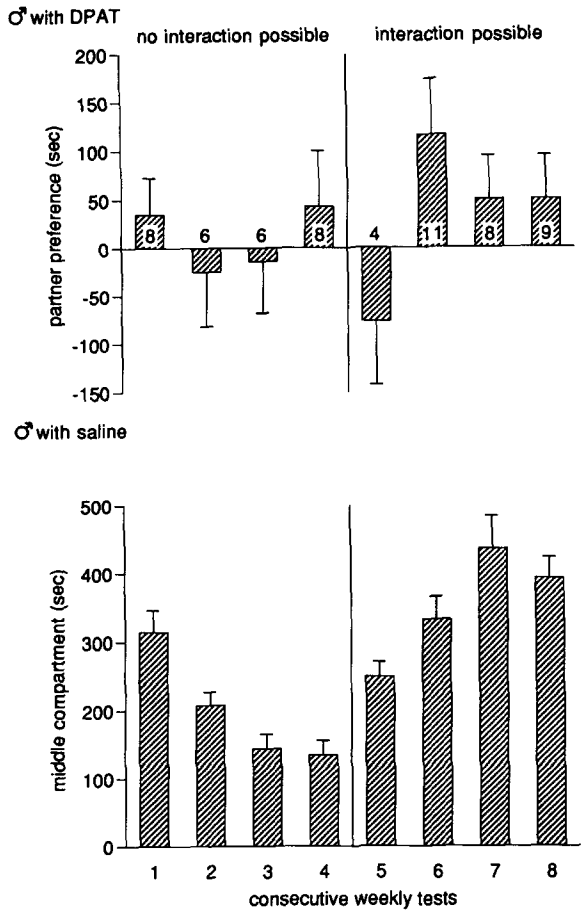


FIG. 2. (Top) Mean (±SEM) preference (seconds) for a 0.2 mg/kg 8-OH-DPAT-treated male over a saline-treated male of estrous female rats (n=15), tested in a 3-compartment box (3CB; 15 min/test). During tests 1–4 behavioral interaction was prevented by a wire mesh partition, which was removed during tests 5–8. Figures in bars indicate number of females that preferred the vicinity of the 8-OH-DPAT-treated male (i.e., a positive time score). (Bottom) Mean (±SEM) time (seconds) spent in the empty middle compartment of the 3CB. From this also the total time in the lateral compartments may be calculated: total test time adds up to 900 s. For example, test 4: 134 s in middle comp. means 766 s (900 − 134) in lateral compartments.

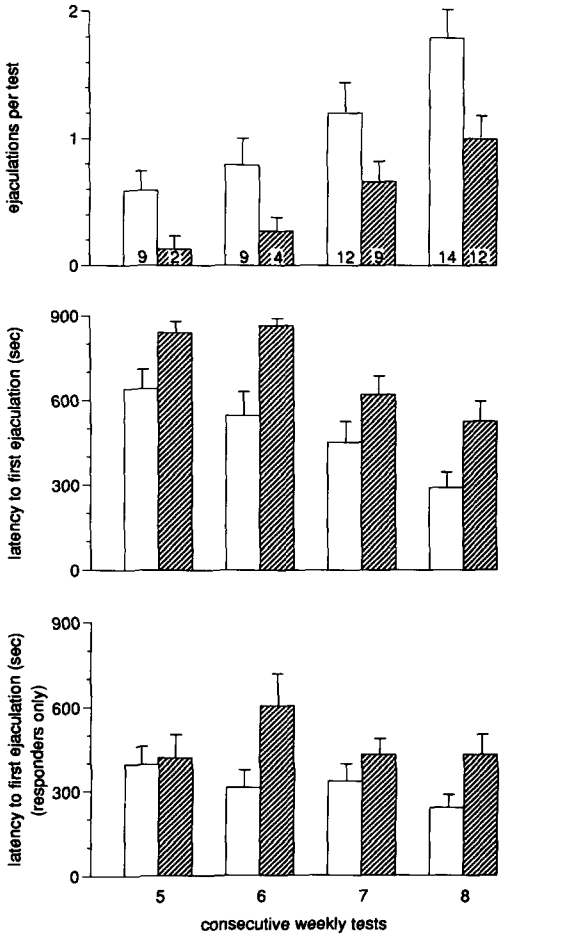


FIG. 3. Various ejaculatory parameters (mean±SEM) of the 8-OH-DPAT-treated (n=15; open bars) and saline-treated (n=15; hatched bars) tethered males during partner preference test 5–8 as depicted in Fig. 2. Figures in bars indicate number of males that ejaculated at least once during that test.

Intromission frequencies (Fig. 4), subjected to two-way ANOVA, differed over the tests, $F(3,42)=5.67$, $p=0.002$, with no overall treatment difference, $F(1,14)=0.21$, n.s., but with a significant test × treatment interaction, $F(3,42)=4.66$, $p=0.007$. This interaction points to the fact that the 8-OH-DPAT-treated males (Fig. 4; open bars) had somewhat similar mean intromission frequencies over the 4 tests (range: 2.3–3.7), while the saline-treated males (hatched bars) showed an increase over the tests (range: 1.2–5.7). Mount frequencies subjected to ANOVA showed a significant effect of tests, $F(3,42)=5.17$, $p=0.004$, no effect of treatment, $F(1,14)=0.41$, n.s., and a test × treatment interaction, $F(3,42)=2.75$, $p=0.05$.

In summary, in the course of testing there is an overall increase in copulatory activity of the females with the tethered males. With regard to the number of ejaculations and latency to first intromissions, the 8-OH-DPAT males are more preferred and allowed to be more sexually active.

## EXPERIMENT 4: DOES 8-OH-DPAT STIMULATE MOUNTING BEHAVIOR IN FEMALE RATS?

8-OH-DPAT has stimulating properties in male rats for sexual behavior, and more specifically ejaculation behavior [(1,25); see also Experiment 2 of the present study]. Female rats with and sometimes without exogenous testosterone readily mount an estrous conspecific [e.g., (5,6)]. The question was investigated whether or not 8-OH-DPAT also had "aphrodisiac" properties in female rats.

### METHOD

Seventeen heterosexually naive female rats, approximately 3 months old, were ovariectomized. Three weeks later behavioral testing started, i.e., a 15-min pair-test with an estrous female. Testing occurred twice weekly, 3 or 4 days apart. Animals were tested in three series of 7 tests each. In each test series these 7

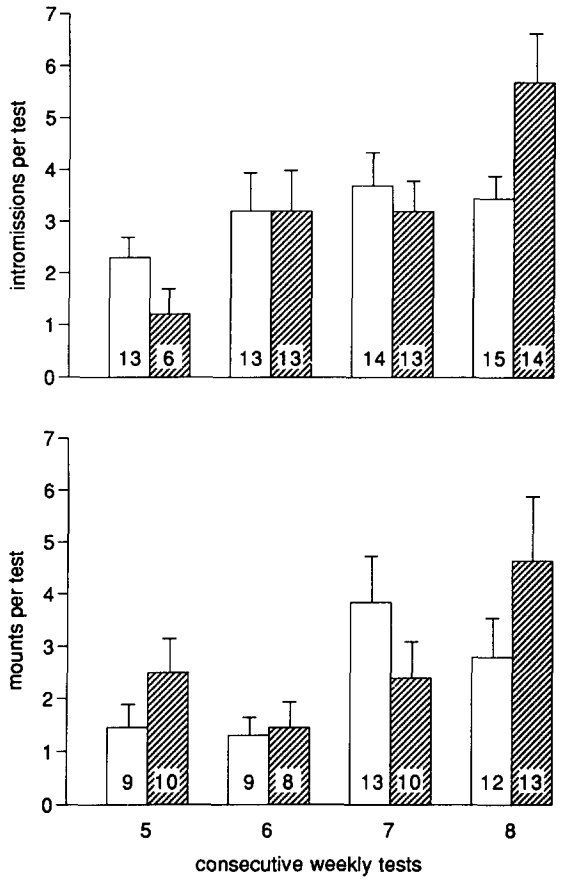


FIG. 4. Intromissions (top) and mounts (bottom) (means ± SEM) of the 8-OH-DPAT-treated (n = 15; open bars) and saline-treated (n = 15; hatched bars) tethered males during partner preference test 5–8 as depicted in Fig. 2. Figures in bars indicate number of males that showed the behavior.

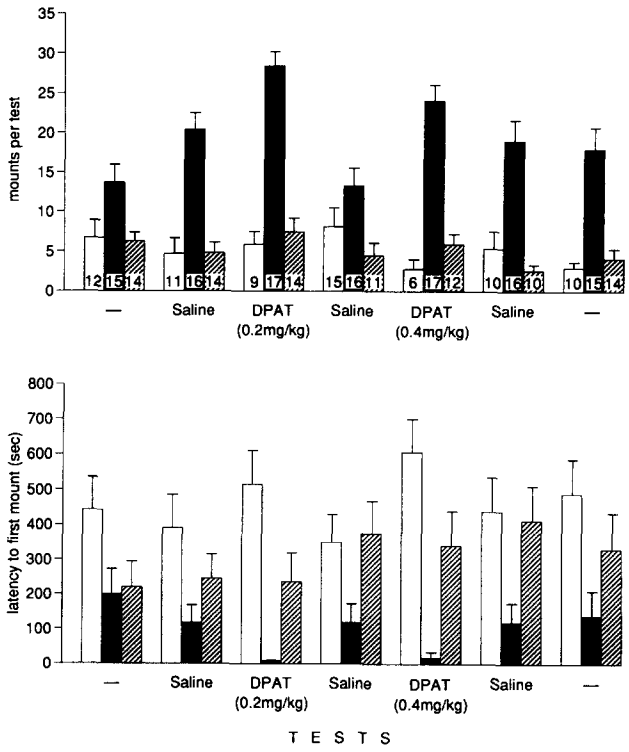


FIG. 5. Mounts and latency to first mount (mean ± SEM) of female rats (n = 17) during 3 test series: before (open bars), during (black bars) and after (hatched bars) long-term testosterone treatment. Each test series comprised 7 tests (15 min/test; twice weekly) in a fixed sequence of treatment: none, saline, 0.2 mg/kg 8-OH-DPAT, saline, 0.4 mg/kg 8-OH-DPAT, saline and none.

tests comprised a first and a last test without any exogenous treatment and tests 2 through 6 with SC injections with saline, 8-OH-DPAT (0.2 mg/kg), saline, 8-OH-DPAT (0.4 mg/kg) and saline, respectively.

Following the first test series each female received SC in the neck under light ether anesthesia a silastic implant filled with testosterone (inner diameter 1.5 mm, outer diameter 2.1 mm, effective length 2 cm). Three weeks later test series 2 was started. One week following the last test of the second test series the implants were removed and the females were left undisturbed for 7 weeks. Then the final test series were carried out.

During behavioral tests mounts with pelvic thrusts and intromission-like mounts were scored, and latency to first mount was recorded.

## RESULTS

Mount frequencies and latencies to first mount are delineated in Fig. 5. The data were subjected to two-way ANOVA [data were normalized by taking the square root of each of the data (15)] with factors testosterone and saline/8-OH-DPAT treatment. For mount frequencies there was an effect of testosterone, $F(2,320) = 152.5$, $p<0.001$, of saline/8-OH-DPAT treatment, $F(6,320) = 2.70$, $p<0.01$, and a significant interaction, $F(2,320) = 3.12$, $p<0.01$. Analysis with the simple main effects method (15) only showed a significant overall effect during testosterone treatment, $F(6,320) = 5.85$, $p<0.001$; test series 1 and 3 did not differ, $F(6,320) = 1.60$, n.s. and $F(6,320) = 1.50$, n.s., respectively. Subsequent analysis with LSD method [LSD(5%) = 1.59 for transformed data] revealed that the highest levels of mounting behavior were displayed during 8-OH-DPAT treatment.

Transformed (square root) data for latency to first mount subjected to two-way ANOVA, only showed a significant effect of testosterone, $F(2,32) = 35.45$, $p<0.001$, no effect of tests, $F(6,96) = 0.83$, n.s., and no significant interaction, $F(12,192) = 1.69$, $p = 0.07$. LSD analysis of the data during testosterone [LSD(5%) = 4.82 for transformed data] showed shortest latencies to mount with 8-OH-DPAT treatment.

Occasionally females displayed intromission-like behaviors. Very low frequencies were seen in tests without 8-OH-DPAT in all 3 test series (range 0–3 "intromissions" in 17 females). 8-OH-DPAT treatment increased the number of females displaying this behavior: low dose 8-OH-DPAT 3–5 out of 17; the higher dose 8-OH-DPAT 6–11 out of 17. The highest frequency, 11 of 17 females, was observed in the testosterone condition (i.e., test series 2).

## GENERAL DISCUSSION

The main findings of the present experiments can be summarized as follows. 8-OH-DPAT treatment stimulated ejaculation frequency in "middle-aged" (approximately 15 months old) male rats, both in initially sexually active and in initially sexu-

ally inactive subjects. 8-OH-DPAT made both groups equally sexually active. The increased number of ejaculations per test coincided with a decrease in total number of mounts, of intromissions, of intromissions prior to first ejaculation and of latency to first ejaculation.

In the second experiment 8-OH-DPAT rendered a high percentage (45–55%) of males to ejaculate "prematurely," i.e., at the first or second intromission. Latency to ejaculation decreased. With the higher dose 8-OH-DPAT (0.4 mg/kg b.wt.) 25–35% of the males ejaculated extravaginally.

8-OH-DPAT treatment did not make males more attractive for an estrous female, as judged by the time spent in the neighborhood of such males. The estrous females received far more ejaculations from the tethered 8-OH-DPAT-treated males, with the lowest latencies to first ejaculation, than from the saline-treated males.

In the final experiment it was found that 8-OH-DPAT treatment stimulated mounting behavior in female rats only when they were long-term treated with testosterone. During testosterone treatment the shortest latencies to first mount were found after 8-OH-DPAT administration.

The facilitatory effects of 8-OH-DPAT on male sexual behavior and sexual motivation as judged by latency to ejaculation in our middle-aged male rats are similar to what has been found earlier in younger rats (2, 21, 25). The stimulatory effects of 8-OH-DPAT in sexually active and inactive middle-aged gonadally intact male rats seem to be a new finding.

The premature character of the ejaculatory behavior following 8-OH-DPAT treatment is in line with other studies (2,21). Not reported in the literature is the finding that relatively many males (25–35%) ejaculated extravaginally. Two other studies that collected and examined the coagulated ejaculate or copulatory plugs reported an "impairment in ejaculation" (25) or "a trend towards weight reduction" (18). These conclusions were based on lower plug weights with 8-OH-DPAT.

The mechanism through which the 5-$HT_{1A}$ agonist 8-OH-DPAT facilitates various aspects of ejaculatory behavior is not yet clear. It could potentially affect animals via both central and peripheral pathways. Finberg and Vardi (11), studying the inhibitory effects of 5-HT on penile erectile function in the pithed rat, found that 8-OH-DPAT had no effect on intracavernous corporal pressure in the penis. This suggests that 8-OH-DPAT does exert its effect centrally rather than peripherally. The present results with the testosterone-treated females (Experiment 4) also point towards a central action of the drug.

Although 8-OH-DPAT treatment stimulates sexual behavior in males, it does not make them more attractive to estrous female conspecifics. When given the choice between 8-OH-DPAT-treated and saline-treated males, females have no preference for either one as judged by the time spent in their vicinity. It does not make a difference whether or not sexual behavior with the stimulus males is possible. When sexual behavior is possible, females copulated with both males but spent most of the test-time in the neutral compartment. This contrasted with the situation in which sexual interaction was prevented by a wire mesh partition. Then, females spent most of their time in proximity of either male. It was earlier found that estrous females prefer a castrated male over a gonadally intact male (7,9). Such preference disappeared when the vagina was taped, which prevented intromissions and ejaculations to occur (7). Although in the present experiment 8-OH-DPAT-treated males had lower intromission frequencies to ejaculation than controls this did not make them more attractive. This can be explained by the fact that the total intromittive activity was virtually similar for both stimulus males. In a recently published paper flesinoxan, just as 8-OH-DPAT a 5-$HT_{1A}$ agonist, increased ejaculation frequencies in tethered male rats and made them more attractive to estrous females (23). This latter finding differs from the present results. There are many differences in the testing procedures between the two studies. For instance, Mos et al. administered different drugs (a total of 5) or vehicle in a cross-over design to each of 8 pairs of male rats, with intervals between the tests of 3 or 4 days. It remains to be tested whether flesinoxan renders tethered males more attractive when animals are tested repeatedly as was done in the present study.

In the present study, the facilitatory effects of 8-OH-DPAT on mounting behavior in female rats were only present with concomitant long-term testosterone treatment. Also 8-OH-DPAT enhanced sexual motivation in testosterone-treated female rats, as judged by latency to first mount. These data corroborate the preliminary results of Mendelson and Gorzalka (21). In the latter study females were testosterone treated for 3 weeks (SC 100 μg TP/day) and tested only once for masculine sex behavior. Although in the present study it was investigated in much more detail (repeated twice weekly tests, before, during and after testosterone treatment), basically the same results were obtained.

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