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Opiate Antagonists and Sexual Behavior in Female Hamsters[1]

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OSTROWSKI, N. L., R. G. NOBLE AND L. D. REID. *Opiate antagonists and sexual behavior in female hamsters.* PHARMAC. BIOCHEM. BEHAV. 14(6) 881–888, 1981.—Mating or administration of morphine to female hamsters reliably decreases lateral displacement, a sensitive index of female sexual responsivity. Morphine's effects are antagonized by naloxone. We asked whether endogenous opiates are significantly involved in the mating-induced inhibition of sexual responsivity by testing whether naloxone or naltrexone attenuated the mating-induced decreases in lateral displacement. Naloxone (4 mg/kg) increased lateral displacement in only one of three tests in females before mating. Naloxone did not attenuate the mating-induced decreases in lateral displacement or lordosis behavior in either ovariectomized, hormonally supplemented or intact females. Large doses of naltrexone produced no reliable effects on sexual behavior during estrus in unmated females, nor did it attenuate the mating-induced decreases in sexual responding, regardless of the time of day of mating. Naloxone often increases the variability of sexual responding. We conclude that naloxone-sensitive mechanisms do not play a critical role in the expression of sexual behavior in female hamsters.

Sexual behavior Hamsters Naloxone Endorphins Opiates Female

MATING reduces sexual responsivity and abbreviates the duration of behavioral estrus in a variety of mammals [2, 8, 15, 16]. Although the specific mechanisms responsible for these phenomena have not been detailed for any species, the Syrian hamster has been studied in attempts to characterize the mating-induced inhibition of sexual responsivity [6, 7, 8]. Apparently, vaginocervical stimulation, independent of chemicals contained in the male's ejaculate, is necessary and sufficient to produce abbreviation of behavioral estrus in hamsters [7]. Also, attenuation of sexual responsivity after mating has been observed in intact as well as ovariectomized and hormonally supplemented females and in adrenalectomized and hypophysectomized females [6,8]. These results lead to the suggestion that the mating-induced inhibition of sexual responsivity is not mediated entirely by these endocrine mechanisms.

Exogenous opioids inhibit sexual behavior and impair endocrine and reproductive functioning in male and female rats [4, 5, 9, 10, 30, 31, 32, 39] and in human beings [22,41]. A reasonable suggestion is that release of endogenous opioids during mating might be part of the sequence of events leading to the mating-induced suppression of sexual responding.

The idea that endogenous opioids may be involved in sexual functioning after mating is further supported by the finding that plasma β-endorphin-like immunoreactivity is elevated as much as 86-fold in male hamsters immediately after ejaculation [25]. Also, blockade of opiate receptors with naloxone or naltrexone produces changes in male copulatory behavior which have been reported to resemble facilitatory effects on components of sexual behavior [14, 18, 24, 36]. This series of experiments follows our previous work [29] which demonstrated that morphine suppressed components of female hamster sexual behavior.

Lateral displacement is the female hamster's response to tactile stimulation of the perineal region by laterally moving the vaginal midline toward the stimulation [28,29]. This hormone-sensitive sexual response by the lordotic female facilitates intromission by the male and can be easily quantified [28]. Morphine administration and mating both produce dramatic reductions in lateral displacement within 15 min [28,29]. The morphine-produced decreases can be reversed by naloxone [29]. We now ask whether naloxone can modify the mating-induced decreases in this, and other sexual responses in female hamsters. We also report here

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effects of variables, such as time of testing during estrus and hormonal state, on lateral displacement and lordosis behavior.

EXPERIMENT 1

If endogenous opioids are involved in the regulation of female sexual behavior, then blockade of opiate receptors would be expected to alter sexual performance. This experiment was conducted to determine whether naloxone attenuated the mating-induced inhibition of sexual responsivity in female hamsters.

METHOD

Subjects

Twenty, sexually experienced female hamsters (*Mesocricetus auratus*), weighing about 120 g, were used. Animals were purchased from Engle's Laboratory Animals, Inc., and housed in groups of 4 or 5 in solid-bottomed, plastic cages (38×33×17 cm) on corn-cob bedding. Animals were maintained on a reversed light-dark cycle (14 hr of light, 10 hr of dark with lights off at 1400 hr). Sexually active male hamsters were used as stimulus males in tests of sexual behavior. Standard laboratory chow and water were always available in home cages.

Females were ovariectomized at least one week before testing. At the time of surgery (performed under sodium pentobarbital anesthesia) each female received a subcutaneous (SC) implant containing 17-β-estradiol in crystalline form. The implants (15 ga metal tubing, 1 cm long) were capped with Silastic and had been calibrated to provide constant circulating levels of estradiol which approximate those observed in proestrous females [27]. Each female was injected with 0.50 mg of progesterone, SC, dissolved in 0.10 ml of corn oil, 4 hr before the beginning of testing to induce behavioral receptivity.

Apparatus

Apparatus for all experiments included several 5.5 gal glass aquaria which were used in tests of sexual behavior. A set of Semmes-Weinstein aesthesiometers (Stoelting Co.) was used for applying perineal stimulation during tests for lateral displacement. Either a set of 11 probes (pressures ranging from 66.8 G to 0.69 G of force) or an abbreviated set of 4 probes (pressures ranging from 20.7 G to 1.83 G of force) was applied in descending order of pressure. The magnitude of lateral displacement scores from experiments where the 11 probe set was used is greater than scores from experiments where the 4 probe set was used.

Procedure

Females were randomly assigned to one of two groups (n=10/group) to receive either 4 mg/kg of naloxone HCl or a similar volume of 0.9% saline, SC. This dose was selected on the basis of previous work [29]. Testing began between 1400 and 1600 hr and was conducted in a well-illuminated room by individuals unaware of drug treatment. Females were tested for lateral displacement before drug injection (baseline or Base), 15 min after injection (post-injection or P-I) and then animals were mated. During mating each female received 40 intromissions, including ejaculations, within 15 min. Females were tested for lateral displacement again at 5. 15, 30, 45 and 60 min after the last intromission of the mating session (post-mating or PM).

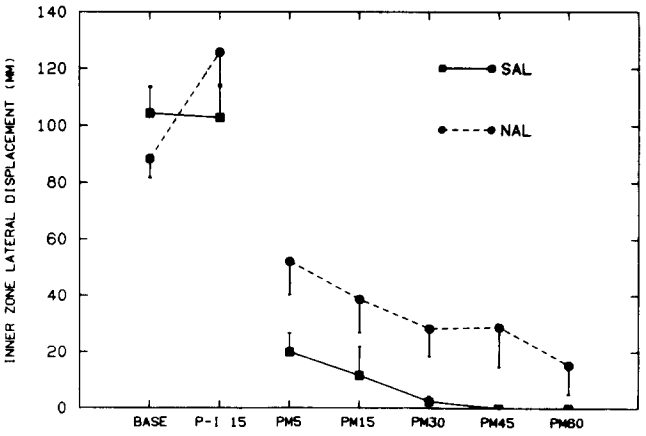

FIG. 1. Effects of naloxone on lateral displacement before and after mating. Group means and standard errors are presented for animals receiving 4 mg/kg of naloxone (n=10) and animals receiving saline (n=10). Animals were tested before and after injection (Base and P-I 15, respectively) and then mated. Animals were retested 5, 15, 30, 45 and 60 min post-mating (PM). Naloxone increased lateral displacement before mating but failed to attenuate the post-mating decreases in lateral displacement, although animals maintained the relative increase in response magnitude throughout tests. Group means differed at 5 and 30 min post-mating ($ps<0.05$, t-tests for comparing means using corrections for unequal variances, [41]), when there were also increased variance estimates relative to the control group (F-tests, [41]).

In tests for lateral displacement, a sexually active male hamster was placed in the aquarium with the female and permitted to investigate but not to mount the female. After the female assumed lordosis, the experimenter applied light tactile stimulation to the back and flanks sufficient to maintain the lordosis for an extended period. While the hamster was in lordosis, the experimenter also applied each Semmes-Weinstein aesthesiometer to the area surrounding the vagina. The probes were applied in a pulsatile manner to the left and right outer zones (±0.7 cm from the ano-vaginal midline) and to the left and right inner zones (±0.2 cm from the ano-vaginal midline). The amount of lateral movement of the vaginal midline in response to each stimulus was measured in mm. When the female failed to respond to two consecutive pressure applications, the test was terminated.

Since inner zone sums of lateral displacement (the sum of each female's responses to the series of stimuli applied to the left and right inner zone) are a sensitive index of female sexual responsiveness and correlate well with other derived measures (rho>.90), they are reported here. Females that did not show lordosis during a test were assigned a score of 0 for purposes of statistical analysis.

Statistical Analysis

Analyses of variance (ANOVAs) having a factor for drug condition and having repeated measures corresponding to repeated tests were used. Separate ANOVAs were also used to determine whether naloxone affected lateral displacement before and after mating. Simple main effects were tested using t-tests and using corrections for heterogeneity of variance, when appropriate [41]. Variance estimates were compared between groups using F-tests [41]. Differences between the numbers of females remaining receptive during tests were determined using Fisher's exact probability tests.

RESULTS

From inspection of mean lateral displacement scores (Fig. 1), it appears that naloxone leads to an increase in lateral displacement before mating and this increase in lateral displacement is carried over to the post-mating tests. A 2 by 7 factorial ANOVA of the scores used to derive Fig. 1, having one factor associated with drug condition and one factor associated with repeated tests, indicated that the naloxone group's scores were greater than the saline group's scores, $F(1,18)=5.54$, $p<0.03$. Also, there was a reliable effect across repeated tests, $F(6,108)=61.7$, $p<0.001$, as expected, since mating intervened between two of the tests (P-I 15 and PM5, Fig. 1). The group by repeated test interaction was also a reliable effect, $F(6,108)=2.25$, $p<0.05$.

Naloxone's Effects Before Mating

To determine whether naloxone facilitated lateral displacement before mating an ANOVA of only pre-mating scores was done (a 2 by 2 ANOVA having factors for drug condition and for tests before and after injection). A reliable group by repeated test interaction, $F(1,18)=13.74$, $p<0.002$, was the only reliable source of variance. There was a reliable increase in responding of the group receiving naloxone (t-test for dependent scores, $t(9)=3.92$, $p<0.01$, while the saline subjects responded similarly on both tests, $t(9)=0.34$, $p>0.50$.

Naloxone's Effects After Mating

Not all females remained receptive for all post-mating tests. Forty-five min after mating, one saline-treated and six naloxone-treated females assumed lordosis ($p<0.05$). One hour after mating, the corresponding numbers were zero and five ($p<0.05$). Direct comparisons of responding post-mating indicate that naloxone- and saline-treated animals differed reliably on scores of lateral displacement at 5 and 30 min after mating (ps<0.05, t-tests for comparing means of samples having unequal variances). At these times naloxone-treated subjects showed reliable increases in the variability of responding.

The higher scores of the naloxone group before mating were maintained during post-mating tests. Consequently, it is questionable whether the apparent increase in post-mating performance in the naloxone group reflects naloxone effects after mating or is an artifact of the increase observed pre-mating. To assess this possibility, a 2 by 2 ANOVA of naloxone- and saline-treated subject's scores obtained pre-mating (P-I 15) and scores obtained 5 min after mating (PM5) was done. Seventeen of the 20 subjects remained receptive for this post-mating test. Mating produced a reliable reduction in lateral displacement as confirmed by the reliable repeated test effect, $F(1,18)=59.4$, $p<0.001$. Naloxone-treated subjects showed greater lateral displacement than saline-treated subjects, $F(1,18)=6.86$, $p<0.02$, however the interaction between group and repeated test was not significant, $F(1,18)=0.20$, $p=0.66$. The absence of a reliable interaction effect suggests that the degree of suppression of lateral displacement produced by mating was similar for naloxone- and saline-treated subjects. This conclusion is further supported by results of a t-test comparing difference scores (scores at PM5−scores at P-I 15) between naloxone- and saline-treated subjects. As can be determined from Fig. 1, the difference in the magnitude of the mating-induced decrease between the two groups is negligible, $t(18)=0.34$, $p>0.50$. Collectively, the results of these analyses provide no support for the hypothesis that naloxone altered the effects of mating on lateral displacement

DISCUSSION

Naloxone increased lateral displacement scores in female hamsters before mating in this experiment. We previously reported a similar facilatatory effect of 4 mg/kg of naloxone on lateral displacement in unmated females [29], but there was also a concommitant increase in the variability of responding. Facilitatory effects of other doses of naloxone were not observed. In subsequent experiments of this report we did not observe reliable increases in sexual responding in unmated females but, occasionally, did find increased variability of responding.

When comparisons of group scores were made taking into account naloxone's effects on premating performance, there was no evidence that naloxone attenuated post-mating decreases in lateral displacement. This is surprising, especially since more naloxone-treated than saline-treated animals continued to assume lordosis during post-mating tests. Because these results leave a number of unresolved issues, the following experiments were done.

EXPERIMENT 2

This experiment was conducted to determine whether naloxone affected indices of sexual behavior other than lateral displacement in female hamsters before mating. We also asked whether naloxone could block the mating-induced inhibition of these measures of sexual responsivity.

METHOD

Subjects

Twelve, sexually experienced female hamsters (Lakeview outbred strain) were prepared as described in Experiment 1, but were individually housed in hanging metal cages on solid bottoms. Animals were handled twice/wk for 3 wk before testing.

Apparatus

In addition to apparatus described, a 2-level chamber (solicitation chamber) which allowed the female to investigate a male, an estrogen-primed female or a neutral area without physical contact was used. The female being tested could freely move across the upper level of the chamber which was separated from the lower 3 compartments by wire mesh (1×1 cm). The lower 3 compartments contained the two stimulus animals which were separated by a larger empty compartment.

Procedure

Animals were randomly assigned to receive either naloxone HCl (4 mg/kg, SC) in 1 ml/kg of 0.9% saline or the vehicle. Behavioral tests were conducted before treatment (Base), 15 min after injection (P-I) and then animals were mated. Mating sessions were terminated after 40 intromissions and did not exceed 15 min. Animals were then tested 5, 15, and 30 min post-mating (PM). The following measures of sexual responding were used. Lordosis latency was the time between the introduction of a male into the test chamber and the assumption of lordosis by the female. Lordosis duration was the time that the female remained in lordosis after re-

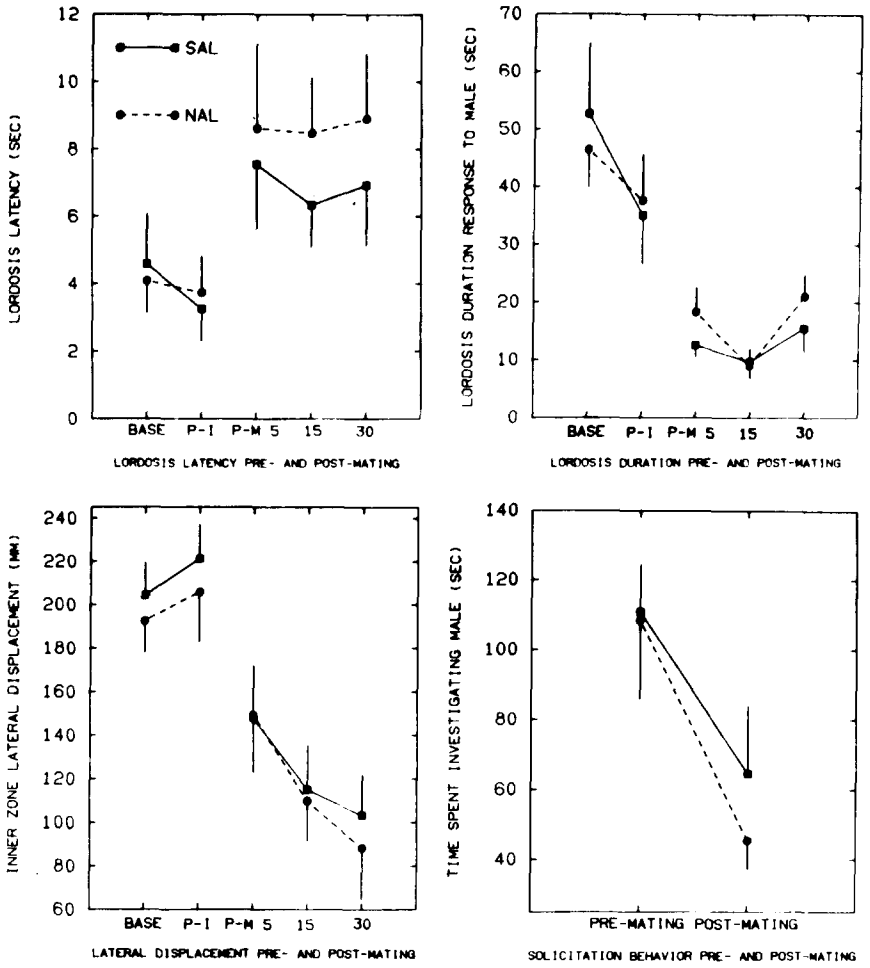

FIG. 2. Effects of naloxone on female sexual behavior. Depicted are means and standard errors for representative data from Experiment 2. Outer zone lateral displacement and post-stimulation lordosis duration are not presented since results are similar to inner zone lateral displacement and lordosis duration in response to a male, respectively. Naloxone failed to increase sexual responding before mating on any index (ANOVAs, ps for data presented >0.50). Naloxone also did not attenuate the post-mating inhibition of sexual responding on any index. All females remained receptive for all tests. Comparisons of group means at each time period indicated that naloxone and saline groups were similar on all indices of sexual behavior (t-tests for independent groups, 17 ps for data presented >0.05). Before treatment=Base, 15 min post-injection=P-I, and 5, 15, and 30 min post-mating=P-M5, 15 and 30, respectively.

moval of the male. Post-stimulation lordosis maintenance was the time the female remained in a lordosis (stimulated by the male's presence) after removal of the male and the termination of 15 sec of light stimulation of the back and flanks of the female provided by the experimenter. Each lordosis response was measured 3 consecutive times and the mean of these scores (sec) was used for analysis. Lateral displacement was measured as described in Experiment 1. Interest in investigating the male, female or neutral area in the solicitation chamber was determined by measuring the time spent over each of the three compartments during a 5 min test. Because of the length of this test, interest in the male was measured only at Base and at 40 min post-mating.

Using similar tests, 41 additional hamsters (Engle's Laboratory Animals) were used in 2 different experiments to assess (a), whether naloxone, when administered during mating, affected post-mating responding, and (b), whether naloxone produced effects on sexual responding in intact females before and/or after mating.

RESULTS AND DISCUSSION

Mating produced reliable inhibition of sexual responding on all measures of sexual responding tested (6 $ps<0.002$), however naloxone neither facilitated sexual responding (6 $ps>0.44$) nor did it attenuate the mating-induced inhibition of responding after mating (6 $ps>0.14$). Comparisons of means between saline-treated and naloxone-treated animals confirmed the lack of a naloxone effect at any time, before or after mating on indices of sexual responding (Fig. 2).

TABLE 1

LATERAL DISPLACEMENT AND LORDOSIS DURATION SCORES AFTER NALOXONE OR SALINE

Experiment	Treatment	Measure	Baseline	Post-Injection 15 min	Post-Mating 15 min	Post-Mating 45 min
A	Saline (n=10)	Lat. Dis. (mm)	134.9 (29.9)	—	96.7 (28.0)	79.5 (32.4)
		Lor. Dur. (sec)	29.1 (6.1)	—	26.0 (5.1)	20.5 (9.3)
	Naloxone (n=11)	Lat. Dis. (mm)	156.5 (19.2)	—	102.6 (23.4)	83.4 (22.5)
		Lor. Dur. (sec)	39.9 (13.7)	—	41.5 (9.2)	28.7 (8.2)
B	Saline (n=10)	Lat. Dis. (mm)	126.4 (16.6)	137.4 (17.5)	125.6 (14.6)	—
		Lor. Dur. (sec)	17.4 (6.9)	26.8 (6.4)	13.8 (2.1)	—
	Naloxone (n=10)	Lat. Dis. (mm)	126.1 (18.5)	130.8 (17.3)	119.4 (20.5)	—
		Lor. Dur. (sec)	17.1 (5.6)	28.3 (9.5)	18.6 (4.5)	—

Presented are the means and SEMs (in parentheses) for two experiments. Experiment A refers to procedures using 21 ovariectomized female hamsters that had been implanted with estradiol containing capsules and also received 6 μg of estradiol 48 hr and 0.5 mg of progesterone 4 hr before testing. Injections of drug or vehicle were made after 12–15 intromissions during the mating session. Mating was terminated after 40 intromissions. Experiment B refers to procedures using 20 intact, regularly cycling female hamsters. Animals received injections of either 4 mg/kg of naloxone or vehicle on their natural day of estrus before mating and the mating session was terminated after 30 intromissions. Naloxone produced no reliable effects on sexual behavior or on the number of females remaining receptive after mating in either experiment. Lat. Dis.=Lateral displacement, mean inner zone sums. Lor. Dur.=Mean lordosis duration.

These data confirm the conclusion of Experiment 1, that naloxone does not attenuate the mating-induced inhibition of sexual responsivity in female hamsters. Unlike results of Experiment 1, however, these results failed to suggest that responding, as indexed by lateral displacement and by lordosis behavior, was potentiated before mating. It is unclear why naloxone increased lateral displacement in one experiment but not in another.

It is possible that the effects of naloxone are short-lasting and potential effects of naloxone on the mating-induced inhibition of lateral displacement might not be easily observed 40 to 50 min after injection, when measurements reported here were obtained. Results of another experiment suggest that this is not a likely possibility (See Table 1-A). Even when naloxone was administered to females about half-way through the mating session, which is less than 22 min before the first post-mating test, naloxone failed to attenuate the mating-induced inhibition of lordosis duration and lateral displacement. These females received injections of 6 μg of estradiol benzoate, 48 hr before testing and 0.5 mg/kg of progesterone 4 hr before testing in addition to the estrogen implants they had received during ovariectomy. Thus, these high estradiol-primed females also failed to show effects of naloxone on post-mating responding, even though nearly all females remained receptive during post-mating tests.

Similar to results reported here for ovariectomized, hormonally supplemented females were results obtained when intact, cycling female hamsters were tested. These females were mated to 30 intromissions to ensure that an adequate number of females remained receptive during post-mating tests. Naloxone failed to potentiate sexual responding before mating and failed to attenuate the mating-induced decreases in lateral displacement and lordosis duration (See Table 1-B).

The results of these experiments lead us to conclude that naloxone does not block the mating-induced inhibition of sexual behavior in females. Additionally, if naloxone facilitates sexual performance in female hamsters, as suggested by results of Experiment 1, it may do so only given special conditions. The following 2 experiments address this issue by (a) testing whether naltrexone, a long-acting opiate antagonist, exerts different effects on sexual responding depending on the time of day of testing, and (b), testing whether naltrexone exerts different effects on the mating-induced inhibition of sexual responding depending on the time of day of mating.

EXPERIMENT 3

Diurnal fluctuations in endogenous opiate levels and corresponding differences in sensitivity to nociceptive stimuli have been reported [12, 13, 40]. We have observed differences in female hamster sexual responding from the beginning to the end of behavioral estrus. We reasoned that if levels of endogenous opiates influenced female sexual behavior, then effects of naltrexone on sexual behavior might be more apparent at times when endogenous opiate levels are higher. For this reason, we tested ovariectomized female hamsters for sexual responding across the day of behavioral estrus (induced by exogenously administered hormones) with or without blockade of opiate receptors.

Since it was important to produces sustained blockade of opiate receptors we chose to use a single high dose of naltrexone HCl. This dose, 100 mg/kg, was selected because by itself, it produced no measurable changes in rearing, locomotion, or responses to nociceptive stimuli in male hamsters (Ostrowski, Noble and Reid, unpublished data), because it antagonized effects of up to 100 mg/kg of morphine on behavioral measures in female hamsters for over 10 hr (preliminary data) and because of recent suggestions that high doses of antagonists might be required to block subtypes of opiate receptors [34].

METHOD

Subjects

Twenty, ovariectomized female hamsters (Engle's Laboratory Animals) were prepared as described in Experiment 1. Females received 0.50 mg/kg of progesterone, SC, 4 hr before the beginning of the first test.

TABLE 2

MEANS AND STANDARD ERRORS FOR LATERAL DISPLACEMENT AT VARIOUS TIMES AFTER NALTREXONE DURING ESTRUS

Group	Hr Post-Injection 2	5	8	11	13
Saline	131.0	130.4	137.9	90.2	50.3*
	(18.5)	(12.0)	(12.2)	(23.1)	(19.0)
Naltrexone	154.0	149.5	141.6	87.2*	62.0*
	(23.3)	(15.9)	(17.8)	(26.7)	(26.4)

Animals (n=10/group) were injected 2 hr before the beginning of behavioral estrus and tested at various times. Means did not differ between groups. The number of females remaining receptive at each time period was similar for both groups. *Indicates scores differ from scores obtained at 2 hr after injection (t-tests for dependent groups, $ps<0.01$). Standard errors are in parentheses.

Procedure

Animals were randomly assigned to receive injections of either 100 mg/kg of naltrexone HCl dissolved in 2 ml/kg of saline or the vehicle, SC. Injections of drug or vehicle were given between 1200 and 1245 hr on the test day. This corresponded to 2 hr after injection with progesterone and 2 hr before the beginning of behavioral estrus (the onset of the lights-off period).

Animals were tested for lordosis latencies and lateral displacement responses as described, at 2, 5, 8, 11 and 13 hr after drug or vehicle injections. Testers were unaware of group assignment.

RESULTS

Results are presented in Table 2 for measures of lateral displacement. The number of females remaining receptive at each time period was similar for both groups. Naltrexone did not produce a change in lateral displacement or in lordosis behavior in unmated females. Although animals showed a reliable decrease in lateral displacement toward the end of estrus, $F(4,72)=21.3$, $p<0.001$, naltrexone did not facilitate lateral displacement, $F(1,18)=0.20$, $p>0.60$ and the drug by repeated test interaction was not a reliable source of variance, $F(4,72)=0.40$, $p>0.80$.

Lordosis latency scores did not differ with repeated tests, $F(4,72)=1.4$, $p>0.20$, with drug injection, $F(1,18)=0.66$, $p>0.40$ and the drug by repeated test interaction was not reliable, $F(4,72)=0.70$, $p>0.50$.

Although there is no evidence to suggest that naltrexone altered sexual responding across behavioral estrus, it might be noted that animals receiving naltrexone showed a slight, non-significant increase in the variability of responding, relative to animals receiving vehicle.

Increases in β-endorphin-like immunoreactivity have been reported for male hamsters after mating [24]. It is reasonable to assume that there may be similar increases in circulating opiates in females after mating. Further, the time of day of mating may determine the extent to which mating can potentiate this release. Naltrexone, then, might be expected to produce different effects on the mating-induced inhibition of sexual responding depending on when, during estrus mating occurs.

EXPERIMENT 4

In this experiment we used intact, cycling females and measured sexual behavior before and after injection of saline or naltrexone, and again after mating, to determine whether

TABLE 3

MEANS AND STANDARD ERRORS FOR LATERAL DISPLACEMENT FOR INTACT FEMALES MATED AT VARIOUS TIMES

Time Mated	Group	Base	Post-Injection	Post-Mating (min) 5	15	30
	Saline	96.6	80.5	65.7	56.3	61.9
1400 hr		(8.4)	(9.7)	(7.6)	(3.5)	(4.9)
(beginning of	Naltrexone	87.0	82.6	45.0	40.7	42.5
behavioral estrus)		(4.0)	(5.8)	(17.2)	(16.3)	(17.2)
	Saline	86.4	83.6	41.3	27.9	25.1
1700 hr		(6.4)	(6.9)	(9.7)	(9.0)	(10.6)
(3 hr into behavioral	Naltrexone	70.7	85.1	41.4	33.0	32.1
estrus)		(10.3)	(11.1)	(23.3)	(18.3)	(17.6)
	Saline	77.6	80.4	31.2	9.1	15.7
2300 hr		(3.6)	(6.8)	(8.1)	(5.7)	(9.6)
(near the end of	Naltrexone	76.7	68.3	8.1	4.8	5.8
behavioral estrus)		(10.7)	(9.7)	(9.0)	(5.4)	(6.5)

Intact female hamsters (n=5/group) were injected with 50 mg/kg of naltrexone HCl or saline after baseline measures were obtained, then mated and tested at intervals after the last intromission of the mating session. Naltrexone did not reliably increase responding before mating and did not attenuate the mating-induced inhibition of sexual responding regardless of the time of day of testing ($ps>0.10$). As expected, animals decreased responding after mating and animals tested toward the end of estrus showed lower levels of responding than animals tested at the beginning and middle of estrus. Standard errors are in parentheses. Data are in mm of lateral displacement to inner zone stimulation.

naltrexone exerted different effects on behavior depending on the time of the testing during estrus and the time of mating.

Subjects

Thirty intact and regularly cycling female hamsters were used. Testing took place on the animal's day of estrus and was conducted using a single-blind procedure. Animals were randomly assigned to one of six groups. Half of the animals received 50 mg/kg of naltrexone HCl dissolved in 2 ml/kg of saline and the other half received only vehicle, SC. Animals were then assigned to be mated at 1400 hr (the beginning of behavioral estrus), at 1700 hr (3 hr into behavioral estrus) or at 2300 hr (near the end of behavioral estrus).

Females were tested for lordosis latencies and for lateral displacement before and 15 min after injection with drug or vehicle, and then they were mated. Mating sessions were limited to 40 intromissions within 15 min. Animals were tested for sexual behavior again at 5, 15 and 30 min after mating. Measures of lordosis latency but not lateral displacement were obtained at 60 min post-mating.

RESULTS

Animals mated near the end of estrus showed a more pronounced decrease in sexual responding than did animals mated at the beginning of estrus ($2\ ps<0.005$). Naltrexone treatment did not affect scores of lateral displacement (Table 3) or lordosis latency before or after mating, regardless of the time of day of testing. The number of animals remaining receptive during post-mating tests was similar for naltrexone and saline groups at each respective time period. Again, naltrexone-treated females showed a consistent, but generally non-significant increase in variance relative to saline-treated animals.

GENERAL DISCUSSION

Mating reduces female sexual responsivity as measured by a number of indices, namely, lateral displacement, lordosis measures and a solicitation measure. As the interval between mating and testing increases fewer females remain sexually responsive. Females are less responsive toward the end of behavioral estrus and mating is more effective in producing decreases toward the end of estrus than at the beginning of estrus. Blockade of opiate receptors by naloxone or naltrexone produces no change in these patterns of sexual responding in ovariectomized, hormonally supplemented or intact female hamsters.

Although naloxone apparently increased lateral displacement before mating in Experiment 1, further attempts to replicate this finding failed to demonstrate a facilitatory effect. Experiment 1 and additional experiments also failed to provide support for the idea that the mating-induced inhibition of sexual responding was sensitive to administration of opiate antagonists.

Trends in the data suggest that naloxone and naltrexone increase the variability of sexual responding indicating that these agents are not inert in this species. It is possible that opiate antagonists are effective in increasing sexual responding in subgroups of female hamsters (e.g., animals demonstrating sub-standard responding at baseline) which is consistent with a report indicating that naloxone facilitated copulatory behavior in male rats which were non-copulators [14]. Slight, inconsistent changes in sexual behavior, such as those reported here, might indicate that opiate antagonists exert effects on processes contributing to, but not essential for the complete expression of sexual behavior. It is also possible that variables not addressed in these experiments, such as levels of estrogen and progesterone, or time of year [11] may influence animal's responsivity to naloxone.

Since only one dose of antagonist was used in each experiment, it could be suggested that the doses chosen were inappropriate. The low dose used in these experiments (4 mg/kg of naloxone) has previously been reported to be an effective dose which antagonized the effects of up to 60 mg/kg of morphine on lateral displacement in female hamsters, and to be the most promising of doses tested in altering lateral displacement in unmated females [29]. The high dose, (100 mg/kg of naltrexone) was effective in antagonizing effects of up to 100 mg/kg of morphine on measures of righting reflex, catalepsy and lateral displacement for considerable lengths of time in hamsters. Further, this dose produced no apparent motor difficulty or behavioral changes in the females tested, nor did naltrexone (10 and 100 mg/kg) alter locomotion or responses to nociceptive stimuli when administered alone to male hamsters (unpublished data). Consequently, it is unlikely that the doses used in these experiments were inadequate in providing antagonism of the well-described opiate receptors, and by inference, their endogenous ligands. It is also difficult to argue that the measures of sexual behavior, most notably lateral displacement, which are sensitive to the influences of mating, time of day and morphine, are not adequately sensitive behavioral measures to index effects of opiate blockade on sexual behavior.

It has been suggested that naloxone is not the most effective antagonist of the analgesia produced by enkephalin and electrical stimulation of the periaquaductal gray region in rats [1]. Opioid-like effects of a compound found in mammalian nervous tissue have been reported to be insensitive to naloxone *in vitro* [3], and non-opiate, non-naloxone sensitive receptors in lymphocytes have been described for B-endorphin [17]. Emerging evidence leads to the suggestion that different sub-types of opiate receptors have different affinities for prototypic antagonists [19, 21, 34]. These findings and the discovery of new endogenous substances with opiate-like effects [20,38] limit the generalizability of negative findings from behavioral tests using naloxone and naltrexone. It is nonetheless surprising, however, that effects of naloxone on sexual behavior are not greater [18, 24, 26, 35, 37], particularly since agonists produce such profound effects on sexual behavior [23, 24, 29, 33, 39].

A preliminary report [4] of naloxone's effects on women showing chronic sexual unresponsiveness indicates that naloxone is also ineffective in modifying these female's responsiveness to erotic stimuli. The authors do point out, however, that there may be a subclass of females that are responsive to naloxone. Additional research is necessary to determine under what conditions endogenous opiates exert effects on female sexual functioning.

The data presented in this paper indicate that naloxone-sensitive mechanisms do not account for the inhibition of sexual responsivity in female hamsters after mating. Likewise, opiate mechanisms do not appear to play a significant role in modulating sexual behavior during estrus. Given the avaialble evidence, it is difficult to conclude that endogenous naloxone-sensitive processes play a significant role in female sexual responsiveness.

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