THE ROLE OF ODOR IN LEARNED AVERSIONS TO COPULATORY BEHAVIOR IN MALE RATS

by

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B. S., Kearney State College, 1981

A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

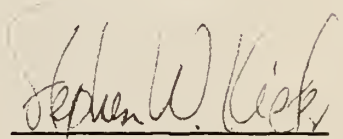
MASTER OF SCIENCE

Department of Psychology

KANSAS STATE UNIVERSITY
Manhattan, Kansas

1984

Approved by



Major Professor

Acknowledgments

I wish to acknowledge and thank the members of my supervisory committee, Drs. Stephen Kiefer, James Mitchell, and Jerome Frieman for their helpful suggestions, support, and encouragement throughout the preparation of this manuscript. A special thank-you goes to Dr. Stephen Kiefer, both advisor and friend, for his advice, guidance, and encouragement not only in the preparation of this manuscript, but also during the entire time he has been my advisor.

I wish to share with my parents the accomplishment of my goals. I thank them for their concern and caring, and for instilling in me an appreciation of education.

I express my gratitude to my dear friend Judy who not only provided constructive criticism of preliminary drafts of this manuscript, but also typed for me and helped me in the literature search. Her constant support and encouragement have been an inspiration to me, and she always gave me a kind word when I needed it most.

Last, but not least, I wish to thank two of my undergraduate professors: Dr. William Wozniak for his demand for detail, excellent teaching, and push to excel; and Dr. Ken Nikels for his direction and endless patience and providing me invaluable laboratory experience.

Table of Contents

Page

Acknowledgments..... ii

List of Figures..... iv

List of Tables..... v

Introduction..... 1

Pilot Study..... 19
- Method..... 20
- Results..... 25
- Discussion..... 31

Experiment 1..... 34
- Method..... 35
- Results..... 38
- Discussion..... 50

Experiment 2..... 52
- Method..... 53
- Results..... 54
- Discussion..... 66

General Discussion..... 68

References..... 76

Appendices..... 87

List of Figures

Figure Page

1. Mean percentage of male rats that displayed ejaculatory responses in the Pilot Study 27
2. Mean latency times for mount, intromission, and ejaculation responses in the Pilot Study . . 29
3. Mean percentage of male rats that displayed ejaculatory responses in Experiment 1 40
4. Mean latency times for mount, intromission, and ejaculation responses in Experiment 1 42
5. Mean latency times for mount, intromission, and ejaculation responses for males that showed an ejaculation response in Experiment 1 . 45
6. Mean number of intromissions to ejaculation by males exhibiting an ejaculation in Experiment 1 . 49
7. Mean percentage of male rats that displayed ejaculatory responses in Experiment 2 56
8. Mean latency times for mount, intromission, and ejaculation responses in Experiment 2 58
9. Mean latency times for mount, intromission, and ejaculation responses for males that showed an ejaculation response in Experiment 2 . 61
10. Mean number of intromissions to ejaculation by males exhibiting an ejaculation in Experiment 2 . 65

List of Tables

Table — Page

1. Treatment conditions of male rats in Experiment 1 36
2. Group sample sizes of males exhibiting ejaculations in Experiment 1 47
3. Group sample sizes of males exhibiting ejaculations in Experiment 2 62

INTRODUCTION

Sexual needs have been thought to be a powerful influence on an animal's behavior, resulting from a strong drive (at least in males) that could not be altered in mammals below the phylogenetic level of humans. As research progressed, observations were made that, except for humans, mammals' mating behaviors were automatic responses dependent only upon an adequate hormone level and proximity to a receptive partner. Although adequate hormone levels are necessary for mating behavior, sexual responses first require an initial stimulus, and in rodents this initial stimulus appears to involve the olfactory system. In fact, the interaction of hormones, olfaction, and sexual behavior appear to be inseparable. However, viewing mating responses as unalterable, automatic behavior is challenged by recent research.

Olfaction

Because of rats' highly developed chemical senses and relatively poor vision, odor stimuli are essential in rats' detecting competitors and predators, food sources, and sexual partners. Nearly from the moment of birth rats learn about and adapt to the environment by receiving olfactory cues from their surroundings. Although the nervous system of a newborn rat is in an embryonic developmental stage (Campbell & Spear, 1972), the dendritic bundles of the olfac-

tory bulbs appear well developed and show a considerable degree of structural maturity (Scheibel & Scheibel, 1975). The relatively large size of the rhinencephalon in the forebrain of the rat reflects the important contribution that olfaction provides to a rat's survival.

Even within the first neonatal day nipple attachment is guided by the odor of amniotic fluid present on the mother (Blass, Hall, & Teicher, 1979; Teicher & Blass, 1977); on ensuing days pups are guided by the odor of milk on the nipples. Rat pups 4-5 days old were shown to stop suckling when the mother's nipples were lavaged to eliminate odors (Teicher & Blass, 1976), and rat pups made anosmic by olfactory bulbectomy failed to suckle (Singh & Tobach, 1975). Singh and Tobach suggested the anosmic pups may not have been able to orient toward the mother and nest area.

Early olfactory imprinting-like processes are important in the control of adult behavior (Johnston & Coplin, 1979). It has been shown (Carter, 1972) that when young guinea pigs were raised with the odor of ethyl benzoate, acetophenone, or no odor, the males later showed more sexual responsiveness toward females scented with the rearing odor than toward unscented females or females scented with the other odor. Whether the process is termed an imprinting process or an associative process in which each odor is indexed to its respective behavioral system (Garcia & Rusiniak, 1980), olfactory imprinting in the neonate may later serve as a

basis for individual and/or species recognition and interaction and play a major role in the survival of the species (Schapiro & Salas, 1970).

The process to recognize and index odors is possible only by the ability of rodents to discriminate between odors. Leon and Moltz (1971) observed odor discrimination in 16 day old rat pups which showed a preference for the odor of their own lactating mother. Lactating rat mothers discriminated between their own and other pups by olfactory cues (Beach & Jaynes, 1956), and adult mice distinguished individual members of the same or different species by olfaction (Bindra & Spinner, 1958). Researchers at Memorial Sloan-Kettering Cancer Center in New York have reported ("Self and Non-Self by Smell," 1979) the discovery of a gene in mice associated with the production of odors that are distinguished by mice. Although the mice distinguished between the odors of urine from mice of same and different genetic types, the only genetic difference was in the major histocompatibility complex gene, the region of the chromosome that contains the genetic information that controls the immunity response.

Olfactory cues are left by both males and females of rodent species by a process known as scent marking; this is accomplished by rubbing the anogenital area over objects in the environment, leaving deposits of urine and preputial secretions (Brown, 1975). Scent marking is a feedback

process to provide the animal with an optimum degree of familiarity with its environment (Eisenberg & Kleiman, 1973), to establish the animal's territory (Pettijohn & Paterson, 1982), and to attract sexual partners (Caroom & Bronson, 1971).

Sexual Behavior

At birth both male and female genotypic rats possess a genital tubercle which is not differentiated into either a masculine or feminine structure (Grady, Phoenix, & Young, 1965). Neural structures that control copulatory behaviors are also undifferentiated at birth (Phoenix, Goy, Gerall, & Young, 1959). Phoenix et al. stated in their organizational hypothesis that androgen is necessary during the first 10 days of life for the masculization of the neural structures responsible for adult male copulatory behavior, and the absence of androgen results in adult display of female copulatory behaviors regardless of the genotypic sex.

Hormone levels in male and female adults are regulated by the hypothalamus which produces releasing factors that stimulate the anterior pituitary to release both follicle-stimulating hormone (FSH) and luteinizing hormone (LH) (Short, 1978). In the female the release of FSH stimulates the Graffian follicle of the ovary to produce and secrete oestradiol-17B (estrogen); this is followed by a marked increase of LH which results in ovulation. Almost coincidental with the LH increase is an enormous preovulatory

surge of progestin (progesterone and 20a-dihydroprogesterone) from the ovary (Short, 1978). The hypothalamus monitors the levels of estrogen and progesterone and directs a 4-day cyclic pattern, if pregnancy has not occurred. Estrous in the female rat is observed only with high levels of both estrogen and progesterone. When estrogen levels drop, either spontaneously or because of an ovariectomy, the female becomes unresponsive to the male (Short, 1978). Receptivity in the ovariectomized female rat can be induced with an injection of 10 µg estradiol benzoate, followed approximately 42 hours later with an injection of 500 µg progesterone (Nikels, 1976).

Receptivity by the estrous or estrous-induced female is portrayed by certain typical behaviors. The most common reaction to genital stimulation is a "courting run" in which the female darts a short distance away from the male, then assumes a posing position with the head pointed slightly upward and the hind feet planted further apart than usual (Beach, 1956). If the male fails to follow, the female approaches the male, often investigating his genitalia, then repeats the darting and pose.

A lordosis response is shown by the female after the male has initiated mating behaviors; lordosis consists of arching the back concavely and moving the tail to one side to expose the vaginal opening. Lordosis response varies from marginal to exaggerated (Hardy & Debold, 1971), and it

is intensified following the male's mount which is accompanied by rapid movements of the male's forelimbs which palpate the female's flanks (Beach, 1956).

In the male, circulating FSH stimulates sperm production, and LH stimulates the testes to produce and secrete testosterone; the testosterone blood level is monitored by the hypothalamus in a negative feedback mechanism to maintain a relatively constant testosterone level (Short, 1978). Sexually experienced male rats do not cease mating behavior abruptly after surgical castration, but show gradually declining sexual behavior for approximately two months at which time sexual activity ceases (Carr, Loeb, & Wylie, 1966). Normal mating behavior can be elicited in castrated males by testosterone injections (Short, 1978).

Mating behavior in the male rat consists of three recognizable responses that include mount, intromission, and ejaculation. A mount consists of the approach from the rear of the female, while the male clasps the female's flanks between his forelegs (Beach, 1956) as the hindquarters move in and out in a series of extremely rapid pelvic thrusts. If penile erection occurs, the male's orientation is appropriate, and the female's lordosis response adequate, a final pelvic thrust results in intromission, the insertion of the penis into the vagina. Penile penetration lasts less than one second and is terminated when the male throws itself vigorously to the rear in a

recognizable "backward lunge" (Beach, 1956) and engages in autogenital cleaning. Intervals from one intromission to the next mount vary from 20 s in some rat strains (Larsson, 1956) to 1 min or more in other strains (Beach & Whalen, 1959; Whalen, 1961). Ejaculation results from a series of intromissions, and the majority of rats intromit 8-15 times before ejaculation occurs (Beach, 1956). Following an initial ejaculation, the male shows no mating response for approximately five minutes and then begins the copulatory sequence again (Beach & Jordan, 1956; Bermant, 1964). In successive copulatory responses to ejaculation the number of intromissions to ejaculation decreases (Larsson, 1956), but the post-ejaculatory refractory time increases (Beach & Jordan, 1956).

Beach (1956) has suggested male sexual behaviors involve two phases and are controlled by two mechanisms. A sexual arousal mechanism is observed as a series of male behaviors that include sniffing the female's head, ears, and anogenital region, pursuit of the female, and physical contact such as crawling over the female. The arousal mechanism is necessary to increase the sexual excitement leading to copulation (Beach & Jordan, 1956).

A copulatory-ejaculatory mechanism mediates the behavioral components of mounting, intromitting, and ejaculation (Beach & Jordan, 1956). A threshold level of sexual excitation is necessary to produce ejaculation, and the threshold

is achieved by a summation of individual intromissions (Beach, 1956; Beach & Jaynes, 1956; Larsson, 1959). Although males may mount and attempt intromission repeatedly, ejaculation does not occur in male rats unable to intromit because of undeveloped penises (Beach & Holz, 1946) or in a situation in which the estrous females have surgically closed vaginas (Kaufman, 1953).

Initiation of activities that result in copulation and ejaculation are displayed by both males and females in all species of mammals (Beach, 1976). Some inexperienced males exhibit incomplete and inappropriate responses when first confronted with a receptive female (Beach, 1956), and the initial activity is begun by the estrous female (Beach, 1942). Inappropriate behavior usually disappears after the male has completed one or two successful copulations. The male that fails to copulate within 5-10 min after the female is introduced is unlikely to do so at that time, even if left with the female for an hour or more (Beach, 1956).

Male rats that ejaculate repeatedly with the same female eventually cease to respond to that female, but resume mating and achieve ejaculation when placed with a different female, a phenomenon termed the "Coolidge Effect" (Wilson, Kuehn, & Beach, 1963). The original female remains receptive and another male may respond to the original female. Maximum sexual behavior in the female is correlated with peak hormone levels, but maximum sexual behavior in the male

varies with the light-dark cycle. Peak activity by the male is seen eight hours after the beginning of the dark cycle, and maximum inactivity occurs 11 hours after the beginning of the light cycle (Harlan, Shivers, & Moss, 1979).

Olfaction and Sexual Behavior

Although adequate hormone levels are both necessary and sufficient (Pfaff, Lewis, & Diakow, 1973) for female receptivity to occur, olfactory stimuli normally summate (Noble, 1973) with tactile sensations and pressure stimuli provided by the male's investigatory and mounting activities (Pfaff et al., 1973) to elicit lordosis. Sexual interactions are known to be dependent on olfactory signals in many species of mammals (Beach, 1976); for example, signalling pheromones in the male mouse preputial gland have been shown to function as a sex attractant (Caroom & Bronson, 1971). Both estrous and diestrous females show discrimination between the odors of gonadally normal and castrated males (Carr & Caul, 1962). Sexually naive (receptive) and sexually experienced (estrous and diestrous) females show a preference for the odor of gonadally normal males; the preference requires either the presence of ovarian hormones or previous sexual experience (Carr, Loeb, & Dissinger, 1965). Reproductive cycles of female mice can even be markedly altered by odors from the male (Parkes & Bruce, 1961) in that priming pheromones from male mouse urine can induce estrous in the female by stimulating release of FSH (Whitten, 1966).

Because estrogen production is closely associated with the timing of ovulation, sexual attractivity of the female to the male is essential to survival of the species because it maximizes the probability of copulation when the female is fertile and susceptible to impregnation (Beach, 1976). Vaginal secretions of estrous females serve as a sexual excitant to adult males (Murphy, 1973), but the attraction is testosterone dependent (Johnston & Coplin, 1979).

Male rats are extremely sensitive to the odor of urine from estrous females, and there is no significant difference between gonadally normal and castrate males in the ability to discriminate the odor (Carr, Solberg, & Pfaffman, 1962). In a study of 12 rats, the least sensitive male discriminated between the odors of air passed over distilled water and air passed over distilled water containing 3 parts per 10,000 of urine from estrous females (Carr et al., 1962). The most sensitive male showed discrimination between the odors of air passed over distilled water and air passed over distilled water with 1 part per 100,000 urine.

Both gonadectomized and prepuberal male rats can discriminate between the odors of sexually receptive versus non-receptive females (Carr & Caul, 1962; Carr & Pender, 1958; LeMagnen, 1952). Gonadally normal males spend more time investigating both receptive and non-receptive females than do castrate males (Carr et al., 1965), and only normal males show a preference for the odor of receptive females

(Carr et al., 1965; Pfaff & Pfaffman, 1969). Prepuberal or castrate males show no preference for receptive females unless injected with testosterone (LeMagnen, 1952). Sawyer (1981) has suggested castration makes neutral stimuli out of what are normally socially significant odors.

Cells in the hypothalamus respond to stimuli from the olfactory system (Norgren, 1977; Scott & Pfaffman, 1967), and an ejaculatory mechanism may have a locus in the medial preoptic area (MPO) of the hypothalamus. Monopolar electrode stimulation of the MPO is involved with genital reflexes in the male members of opossum (Roberts, Steinberg, & Means, 1967), squirrel monkey (MacLean & Ploog, 1962), and monkey (Robinson & Mishkin, 1966) by producing ejaculation without preliminary copulatory behavior (mount and intromission) in the absence of a female partner. The ejaculatory facilitation in rats has been observed for both the arousal mechanism and the ejaculatory mechanism (Beach, 1956) by a decreased number of mounts and intromissions preceding ejaculation, a shorter latency to approach the female, and decreased refractory times between copulatory series (Malsbury, 1971). Bilateral MPO lesions can produce a complete disappearance of male copulatory behavior without accompanying gonadal atrophy (Lisk, 1967).

Activation of the MPO is greatly influenced by testosterone; the most consistent reactivation of male copulatory behavior after castration results from testosterone implants

in or near the MPO (Lisk, 1967). The MPO may be an integrating area where peripheral stimuli (e.g. odors) and hormone levels interact to trigger the male copulatory-ejaculatory pattern. Neural connections project from the olfactory vomeronasal organs to hypothalamic regions known to influence sexual behavior (DeOlmos & Ingram, 1972).

Detection of odors and neural transmission of olfactory information related to sex appear to be functions of the vomeronasal system. Located bilaterally on either side of the nasal cavity (McCotter, 1912), the paired vomeronasal organs (Jacobson's organs) were thought to be redundant organs in mammals until recently (Wysocki, Wellington, & Beauchamp, 1980). Neural pathways from the vomeronasal and primary olfactory systems have anatomically separate pathways to the olfactory bulbs and higher brain structures (McCotter, 1912). Sex-related odors detected by both the vomeronasal organs and the nasal epithelium summate to achieve the necessary threshold for sexual arousal. Powers and Winans (1975) found mating behavior could be abolished in male rodents by destruction of the vomeronasal pathways alone or in combination with destruction of the primary olfactory system, but that destruction of only the primary system without vomeronasal involvement had no effect on mating behavior. Sawyer (1981) suggested the reason castrate males do not prefer the odor of estrous females, but can discriminate the odor, may be due to a disruption of normal

activity of the vomeronasal system when testosterone is absent. Meredith (1983) found complete removal of the vomeronasal organs without damage to the olfactory system resulted in a significant decrease in copulatory behavior in sexually naive males, but no significant difference in copulatory behavior in sexually experienced males. Meredith suggested the vomeronasal system is critical for inducing appropriate copulatory responses in naive males, but experienced males were able to utilize odor stimuli to the primary olfactory system only.

Modification of Copulatory Behavior

Although mating behavior has been shown to be dependent on olfactory stimuli and adequate hormone levels, it is susceptible to environmental conditioning. When using shock as punishment in avoidance learning in rats, Hayward (1957) demonstrated longer latencies by the males to approach and mount receptive females. While testing male copulatory behavior in environments in which the males had previously been shocked, Beach and Fowler (1959) observed ejaculation occurred with fewer intromissions, and they attributed the change to a "situational anxiety" in which less sexual stimulation was necessary in an animal aroused by stress. Ejaculation with fewer intromissions has been observed also when the intercopulatory intervals were prolonged by the experimenter (Beach & Jordan, 1956; Larsson, 1959; Schwartz, 1956). When rats were allowed only seven intromissions before

the testing period was terminated, males showed longer self-imposed intercopulatory intervals and significantly more sessions in which ejaculation occurred before the seventh intromission (Silberberg & Adler, 1974).

Additional experimental manipulations to modify sexual behavior have centered on attempts to reduce the attractivity of an estrous female by inducing an aversion in the male. It was shown in a pioneering study (Garcia, Kimeldorf, & Koelling, 1955) rats formed an aversion to foods following only one pairing with experimentally-induced illness. The taste aversions were learned despite long delays (Garcia & Kimeldorf, 1957) between the conditioned stimulus of taste and the unconditioned stimulus of illness induced by lithium chloride (LiCl) (Garcia, Ervin, & Koelling, 1966).

Taste has been shown to be a strong cue and odor a weak cue for delayed LiCl poisoning (Rusiniak, Hankins, Garcia, & Brett, 1979) in taste aversion learning. However, odor aversion learning in rats was demonstrated following four odor-illness pairings (Hankins, Garcia, & Rusiniak, 1973); Rudy and Cheatle (1977) demonstrated odor aversion learning in rat pups only two days old. Ten-day old rat pups showed an odor aversion to apple juice to which they had been exposed and made ill as fetuses on gestation day 20 (Smotherman, 1982).

It has been suggested (Garcia & Ervin, 1968) the close anatomical relationship of afferents for taste and afferents

from the gastrointestinal tract may be responsible for the rapid acquisition of taste aversions following poisoning. Taste and gastrointestinal afferents converge in the brainstem in the nucleus solitarius, and an "emetic center" (Borison & Wang, 1953) is located just lateral to the nucleus solitarius in the lateral reticular formation. However, the anatomical location of the odor afferents provides the unique role of olfaction in arousal of social and reproductive behavior. Because olfaction has primarily a telereceptive function, it may not possess the properties necessary for one trial odor-illness aversion learning (Hankins et al., 1973).

Because previous studies had allowed animal subjects to drink fluids during training and testing periods and had reported odor aversion learning by the suppressed ingestion of fluids, Domjan (1973) suggested odor aversions were possible only for ingestive behaviors. By testing odor aversion learning in rats in the absence of ingestion, Domjan found odor aversion learning did occur, and the rats exhibited decreased exploratory behavior in the presence of odors associated with poisoning.

In an attempt to modify sexual behavior by employing aversion learning, Emmerick and Snowdon (1976) established an aversion in male hamsters to phenylacetic acid, then placed the males with a receptive female swabbed with phenylacetic acid. The males showed increased latency to mount

and decreased time in anogenital sniffing, but no other changes were seen in the male's mating behavior. Johnston and Zahorik (1975) exposed sexually naive male hamsters to a glass plate smeared with vaginal secretions from an estrous female, and induced LiCl illness after the males licked the plate. After a single exposure, the subject males showed a strong taste-odor aversion to the secretions, but no testing was performed to observe subsequent sexual behavior. Expanding on the Johnston and Zahorik study, a follow-up report (Johnston, Zahorik, Immler, & Zakon, 1978) showed a strong taste-odor aversion and noted longer latencies to initiate mating behavior when males were placed with a receptive female two days after the conditioning trial. It was suggested (Johnston et al., 1978) mating behavior was not affected by LiCl poisoning unless the poison was paired with the ingestion of vaginal secretions to produce an aversion; the aversion inhibited the arousal mechanism (Beach, 1956) by altering the meaning of the vaginal secretions but did not inhibit sexual behavior directly.

Inhibition of sexual behavior in male rats has been demonstrated recently by Peters (1983) who placed male rats with receptive females for 30 min periods, then induced illness in the males with LiCl injections. All males were injected after each trial, non-contingent on the copulatory behavior exhibited during the trial. Gradually increasing latency to initiate copulatory behavior was reported, and

after 5-10 training trials all males ceased to initiate any copulatory behavior. Although no overt aggression toward the female was exhibited, the males kicked at the female when the female approached and showed escape attempts and "agitation" (Peters, 1983) by paw-treading movements. Peters noted the aversion persisted when the males were tested in a novel environment and found no significant difference in aversion learning between sexually experienced and sexually naive males. The aversion to copulatory behavior extinguished after four non-reinforced trials. In a follow-up study, Peters and Blythe (1983) increased the molar concentration of LiCl from 0.15 to 0.3 and found a faster rate of acquisition for aversion learning in rats to copulatory behavior. The male rats retained the aversion for 60 days when allowed no extinction trials.

PURPOSE

Only recently have laboratory experiments on mammals been performed in an attempt to modify mating behavior, and such experiments are few in number. Because olfactory cues are necessary for copulatory behavior, it seems likely that olfactory cues may have been involved with the learned aversions reported by Peters (1983). The complete cessation of initiating copulatory behavior shown by the male rats may have been a result of a learned aversion to the odor of the estrous female. If olfactory cues were involved in the acquisition of the aversion to copulatory behavior, manipulation of olfactory cues should affect aversion learning.

The purpose of the present study was to determine whether male rats show an aversion to copulatory behavior when they have been made ill with LiCl after each ejaculation, and whether the presence of a novel odor on the receptive female became a potentiating cue to facilitate the aversion. It was proposed that aversions would be formed to copulatory behavior and that a novel odor would potentiate the aversion by decreasing the number of trials necessary to establish the aversion and by increasing the number of trials to extinction.

PILOT STUDY

Peters (1983) has shown male rats ceased mating behavior after LiCl illness following each 30 min encounter with an estrous female. The Pilot Study was designed to test inhibition of mating behavior in male rats made ill contingent on an ejaculatory response. Illness was not induced unless ejaculation occurred; this varied from Peters' study in which male rats were made ill whether or not mating behavior responses occurred. One experimental group was exposed to a novel odor on the estrous female to determine whether a novel odor cue would potentiate an aversion to copulatory behavior.

Method

Subjects

Twenty-seven male and eight female Holtzman-derived Sprague Dawley rats were obtained from Sasco, Inc. (Omaha, NE); the males were approximately 60 days old and the females approximately 40 days. Commercial rat lab food and water were available ad libitum; all rats were maintained on a 12-hr light : 12-hr dark cycle. All rats were housed individually upon arrival to the laboratory and given a minimum three weeks' habituation to the laboratory before testing.

Surgery and Estrous Induction

Following the habituation period, all females were ovariectomized after anesthesia was induced with Chloropent (55 mg/kg intraperitoneally). The flanks were shaved and incised bilaterally; the ovaries were externalized, ligated, and removed. At least three weeks were allowed for post-operative recovery. Estrous was induced with an intramuscular (IM) injection of 10 µg estradiol ciprionate 48 hrs prior to testing, and an IM injection of 500 µg progesterone 4-6 hrs prior to testing. All females were screened for receptivity with a stud male immediately prior to each test session; only those displaying vigorous lordosis responses were used.

Apparatus

Testing was conducted in a dark testing room with a

25-watt red light bulb suspended above the testing chamber. The testing chamber was a metal compartment (115 x 72 x 32 cm) partitioned in half with a wooden divider; observation was made from above. An electric timer was used to record latency times.

A standard commercial almond extract diluted with water to a 2% solution was used as the novel odor stimulus. The 2% solution was determined empirically to have no taste to humans. A standard 32-oz spray bottle was used to spray the almond extract solution on the anogenital region of the estrous female. Illness was induced in male rats with an isotonic solution of 0.15 M LiCl in doses of 3% body weight by intragastric intubation. Infant feeding tubes (Pharmaseal, Inc.) were used for the intubations.

Procedure

Male rats were ranked according to weight, then randomly assigned to one of four groups. Copulatory tests were conducted every 4-5 days, and two males were tested concurrently in each half of the testing chamber. Testing was conducted within 4-6 hrs after the beginning of the light cycle, before the male's peak inactivity level occurred (Harlan et al., 1979).

The male was placed into the testing chamber alone for one minute before an estrous female was introduced and timing and observation began. If females were to be odorous, the anogenital region was sprayed with the odor stimulus immedi-

ately prior to her placement into the testing chamber. The spraying was repeated approximately 10 min later; the precise time was chosen in each case so not to interrupt the mating responses. Latencies were timed and recorded to the nearest 5 s for mount interval (the time from the introduction of the female to the time of the first mount), intromission interval (the time from the introduction of the female to the time of the first intromission), and ejaculation interval (the time from the introduction of the female to the time of ejaculation).

Mounts, intromissions, and ejaculations were distinguished on the basis of behavioral criteria (Malsbury, 1971). A mount was recorded only if the male approached from the rear, mounted, and clasped the female's sides. An intromission involved exhibition of a mount and a rapid springing back off the female, followed by autogenital grooming. An ejaculation was distinguished by the prolonged motionless clasping of the female following the final thrust, or by the frozen posture of the male over the female.

The test sessions proceeded until the male ejaculated or until the experimenter terminated the session because of the male's failure to exhibit responses within a set time limit. Termination of the test resulted when the male failed to mount within 10 min, intromit within 15 min, or ejaculate within 20 min. Following each ejaculation, all males were intubated to control for the traumatic experience

of the procedure. Males not made ill with LiCl were given isotonic sodium chloride (NaCl) solution in volumes of 3% body weight via the feeding tube. All intubations were performed within one minute after ejaculation.

Females were counterbalanced so that males were paired with different females on successive tests. Males were counterbalanced so that their position in the testing sequence differed on successive tests, except for the males that were paired with odorous females. The partners of odorous females were tested last in each sequence to avoid contaminating the testing chamber with the novel almond odor.

Midinterval tests were conducted 2-3 days after each copulatory test; each male was placed into the testing chamber for 10 min. The males that had been made ill in the preceding testing situation were intubated and given NaCl in the midinterval tests. Males in the illness/control group were intubated and given NaCl during the copulatory tests, but were made ill during the midinterval tests. The procedures performed during the midinterval tests were conducted to provide controls against (a) possible pharmacological effects of LiCl illness, such as any unspecified illness resulting from LiCl-induced illness, and/or (b) possible aversive conditioning to the test environment.

Males were randomly assigned to one of four conditions. The condition/groups were: (a) sex/ill: Following ejaculation males were intubated with LiCl to induce illness and

intubated with NaCl in the midinterval test, (b) sex/control: Following ejaculation males were intubated with NaCl and were intubated with NaCl in the midinterval test, (c) illness/control: Following ejaculation males were intubated with NaCl and were intubated with LiCl to induce illness in the midinterval test, and (d) odor-sex/ill: Following ejaculation with an odorous female males were intubated with LiCl to induce illness and intubated with NaCl in the midinterval test. If ejaculation did not occur during testing, the males were not intubated. Males were given eight copulation trials; the first ejaculation constituted Trial 1. Nine males failed to ejaculate in five pairings with an estrous female and were omitted from the study; three males died during the study and their data were omitted. Males in the odor-sex/ill group were allowed an extra trial, Trial 9, to copulate with a non-odorous female.

Statistical Analysis

Four 3 x 8 (Groups x Trials) mixed design analyses of variance were performed on latency to mount, latency to intromit, latency to ejaculate, and on the percentage of ejaculation responses. Post-hoc *t* tests were conducted on significant results. If a male failed to mount, intromit, or ejaculate within the set time limits, scores of 600 s (mount), 900 s (intromit), and 1200 s (ejaculate) were assigned for the non-performance.

Results

Because of the wide range of scores (5-1200 s), data were transformed to logarithms (base 10). The raw scores of each latency period and the standard errors of the transformed data are presented in Appendix A. Because 45% of the males were eliminated from the study, results were analyzed for small samples: sex/ill (n=4), odor-sex/ill (n=4), and control (n=7). The two control groups (sex/control and illness/control) showed no significant difference from each other in latency to mount, $F(1,5) = 3.12$, $p > .05$, intromit, $F(1,5) = 2.52$, $p > .05$, or ejaculate, $F(1,5) < 1$, $p > .05$; thus, the data were combined.

The mean percentage of males that ejaculated is depicted in Figure 1. Group effects on the total number of ejaculations approached significance, $F(2,12) = 2.97$, $p < .09$. After the first trial, 25-75% of the males in the odor-sex/ill group failed to ejaculate. Males in the sex/ill group showed a 100% ejaculatory response in all trials except Trials 5-7.

Mean latency times for each group to mount, intromit, and ejaculate are depicted in Figure 2. Significant group effects were found in latency to mount, $F(2,12) = 5.21$, $p < .05$; post-hoc t test indicated both the sex/ill and odor-sex/ill groups differed from the control, but did not differ significantly from each other. Group effects approached significance in latency to intromit, $F(2,12) =$

Figure Caption

Figure 1. Mean percentage of male rats that exhibited ejaculatory responses in the Pilot Study.

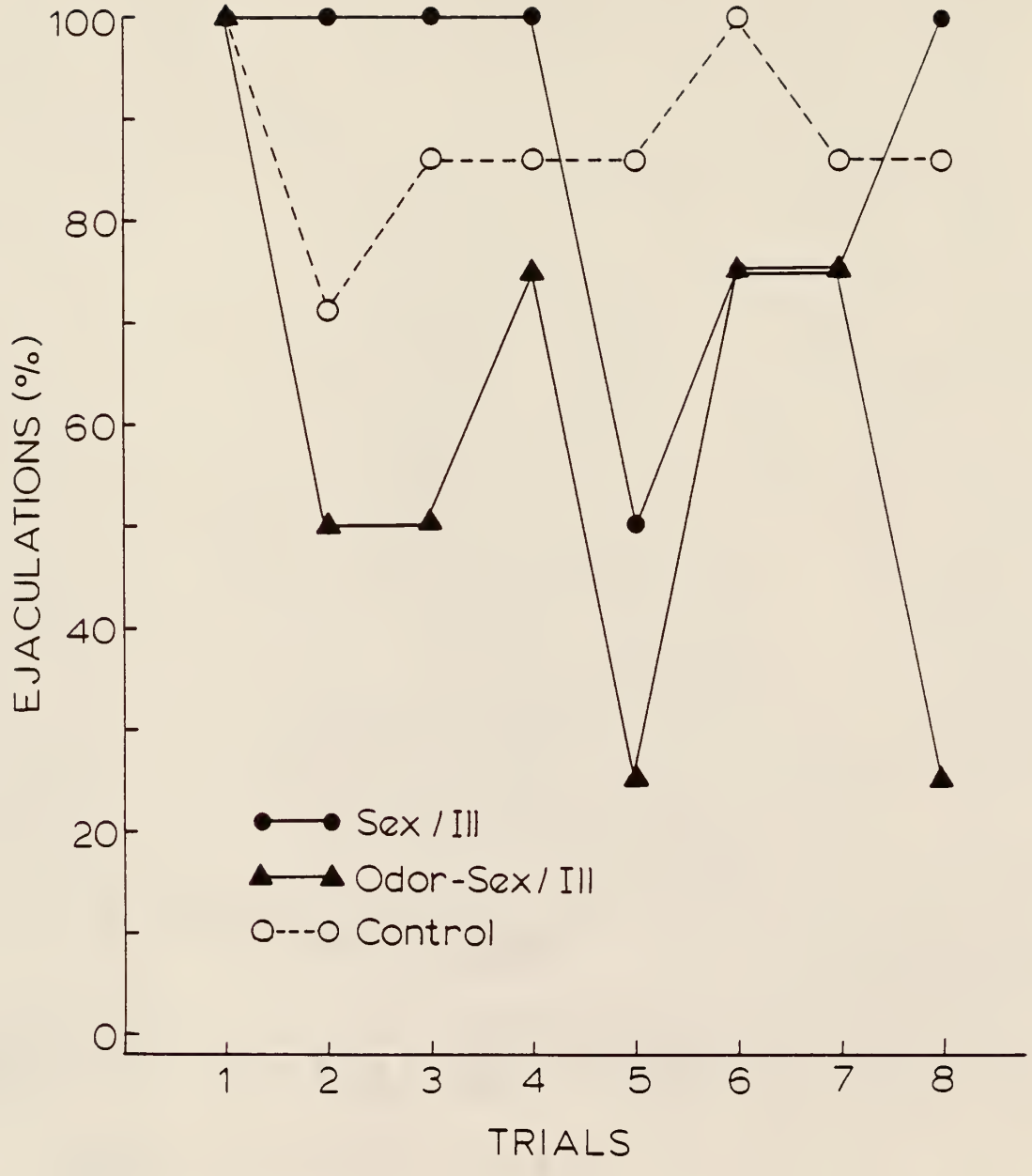
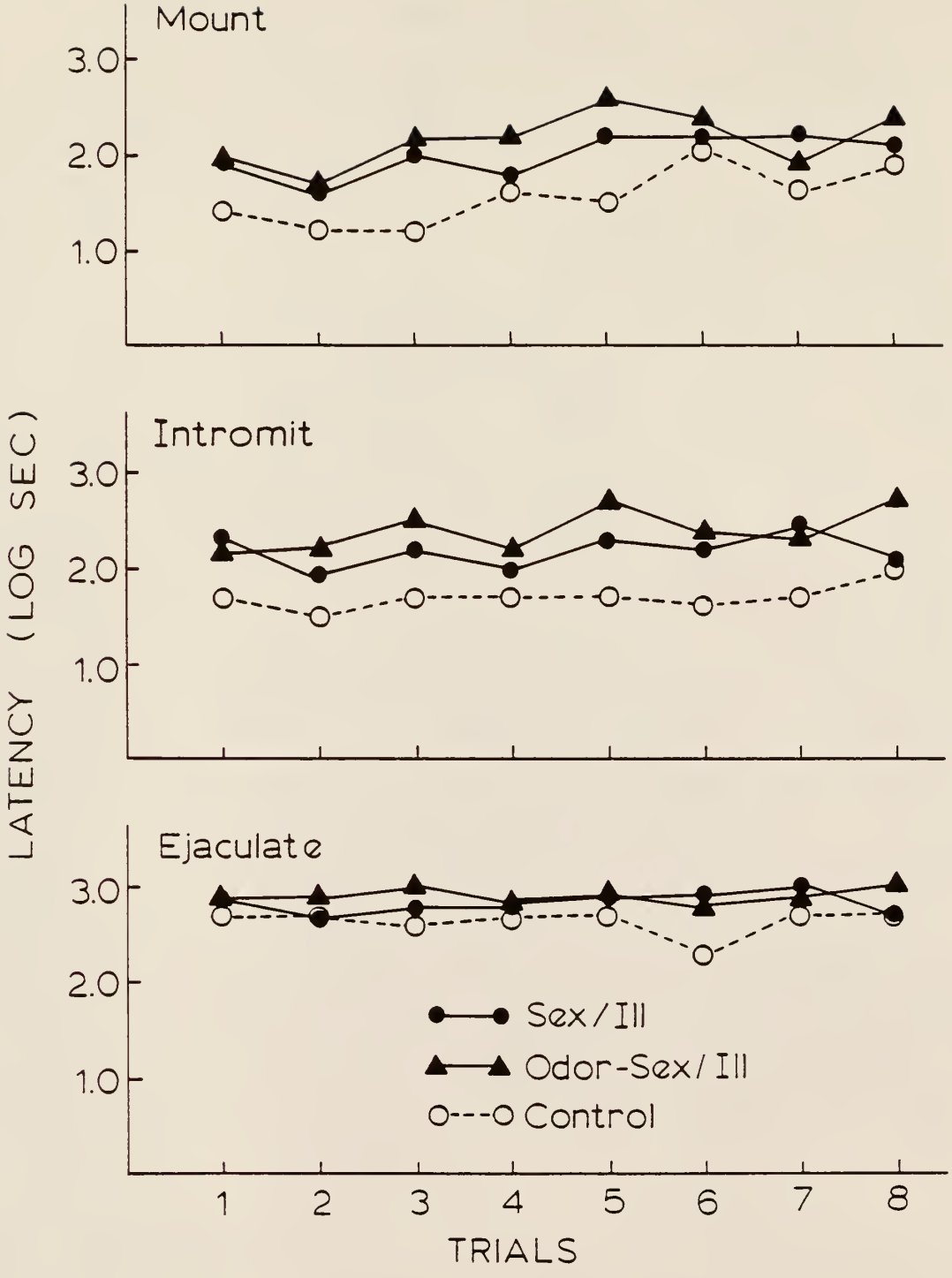

100
80
60
40
20
0
EJACULATIONS (%)
1
2
3
4
5
6
7
8
TRIALS
Sex / III
Odor-Sex / III
Control

Figure Caption

Figure 2. Mean latency times recorded for mount, intromission, and ejaculation responses in the Pilot Study.


Mount
Intromit
Ejaculate
3.0
2.0
1.0
LATENCY (LOG SEC)
Sex/Ill
Odor-Sex/Ill
Control
1
2
3
4
5
6
7
8
TRIALS

3.36, $p < .07$; both experimental groups (sex/ill and odor-sex/ill) showed a tendency for longer latency times than the controls.

Group differences in latency to ejaculate failed to reach significance, but males in both experimental groups showed a tendency for longer latencies to ejaculate than males in the control group. No significant difference was observed in the odor-sex/ill group between Trials 8 and 9 to mount, $t(3) = .93$, $p > .05$, intromit, $t(3) = .21$, $p > .05$, or ejaculate, $t(3) = .40$, $p > .05$.

Discussion

Males in the odor-sex/ill group showed a significant aversion to mount with contingent LiCl-induced illness when compared with the controls. The frequency of ejaculation by the odor-sex/ill group showed marginally significant reductions when compared with the sex/ill group. Each male in the odor-sex/ill group failed to ejaculate on at least two trials, and one male in this group failed to ejaculate in six of eight trials. It is suggested the novel odor cue significantly potentiated the copulatory aversion.

Although not empirically tested, behavior of the males in the odor-sex/ill group was observed to be "submissive." Instead of exhibiting approach responses, the males crouched in the corner of the testing chamber and showed neither aggression nor self-defense behavior even when the estrous female pawed and vigorously bit the male. The only behavior shown by the male in response to the female's biting was an audible vocalization.

During the extra trial (Trial 9), males in the odor-sex/ill group were placed with a non-odorous female. Two of four males showed no copulatory responses; the other two ejaculated at 805 and 1155 s respectively. Comparisons between Trials 8 and 9 failed to reach significance for any of the observed copulatory responses.

Males in the sex/ill group failed to show a significant aversion to copulatory behavior with contingent LiCl-

induced illness. The sex/ill group of the present study received the same treatment as the males in Peters (1983) study, except that males in Peters' study were made ill independent of the copulatory responses. The present study found the sex/ill group required four trials before any male failed to ejaculate, and Trial 5 showed the least number of ejaculations; this corresponds with Peters' observation that 5-10 trials were necessary for a copulatory aversion to occur.

Because two males in the sex/ill group did not ejaculate during Trial 5, they were not made ill, but the same two did ejaculate during Trial 6; it is suggested the aversion extinguished after one non-reinforced trial (Trial 5). One male in the sex/ill group did not ejaculate during Trials 6 and 7, but did so during Trial 8; if this one male had formed a copulatory aversion by Trial 6, then it is apparent the aversion extinguished during Trials 6 and 7 when LiCl was not given. Apparently, a copulatory aversion may require continuous illness reinforcement; males in the Pilot Study were not made ill if ejaculation did not occur, whereas the male rats in Peters (1983) study were made ill during trials following observed aversions to copulation.

Peters (1983) reported males showed an aversion to copulatory behavior by exhibiting no behaviors associated with the sexual arousal mechanism (Beach, 1956), such as approach and investigation of the female. However, the

males in the sex/ill group were observed to mount, intromit, and ejaculate in 93 of 96 (4 males x 8 trials x 3 time-set possible responses) observations, albeit with prolonged latencies. The longer latencies for mount reached significance, but the longer latencies for intromission and ejaculation did not. It is suggested significance was not attained for intromission responses because one male in the sex/control group skewed the means of the control group by exhibiting no copulatory responses in 9 of 24 possible time-set responses.

It is of interest to note that the one control male that exhibited decreased copulatory behavior was in the sex/control group; rats in this group were never given LiCl during copulatory test trials nor during midinterval trials. Other than labeling the rat a non-copulator, it is suggested the decrease in copulatory behavior may be correlated with the fact that all rats were housed individually; this may account for the observation that only 55% of the males exhibited mating behavior. Peters and Blythe (1983) found that 50-75% of the rats raised in isolation showed ejaculation responses; 97% of male rats raised in group colonies showed ejaculatory responses.

EXPERIMENT 1

Experiment 1 was conducted to replicate the results obtained in the Pilot Study with a larger sample size (n=40). An odor/control group was added to test whether the odor itself was aversive to copulatory behavior. Males in the odor/control group were presented estrous females that had been sprayed with the almond odor, but these males were never given LiCl.

Because three subject male rats died during the Pilot Study, the dosage of intragastric LiCl was changed from 3% of body weight used in the Pilot Study to 2% body weight for Experiment 1. In addition, males were housed in groups of five per cage upon arrival to the laboratory to increase the percentage of males exhibiting copulatory responses. A 12-hr light : 12-hr dark reversed cycle was established to test the males during the more active dark phase (Harlan et al., 1979).

Method

Subjects

Forty sexually naive males were grouped five per colony cage upon arrival to the laboratory. After a 3-week habituation period, males were screened 1-3 times for copulatory activity with an estrous female. Males failing to display an intromission response within 30 min of the pre-test trial were eliminated from the study. Following the pre-test trials males were housed individually and assigned randomly to treatment conditions by groups. All male rats were intubated during the test trials following an ejaculation response and during the midinterval trials. LiCl or NaCl was given according to the treatment condition which is summarized in Table 1.

Apparatus and Procedure

Testing was conducted as described in the Pilot Study, except NaCl and LiCl were given in doses of 2% body weight. Copulatory test trials were conducted every 4-5 days during the dark phase of the diurnal cycle. The latencies to mount, intromit, and ejaculate were recorded to the nearest 5 s, and the number of intromissions to ejaculation were recorded. Termination of the test resulted when males ejaculated or failed to exhibit the appropriate responses during the specified time limits. Following ejaculation all males were intubated and given LiCl or NaCl, depending upon the treatment group. Intubations were not performed on males during the

Table 1

Treatment Conditions of Male Rats Following Ejaculation and in the Midinterval Tests in Experiment 1

Group	Test Trial	Midinterval Test
Sex/Ill	LiCl	NaCl
Sex/Control	NaCl	NaCl
Illness/Control	NaCl	LiCl
Odor-Sex/Ill	LiCl	NaCl
Odor/Control	NaCl	NaCl

copulatory test trials if ejaculation had not occurred.

Only males that showed an intromission response during the pre-test trials were used in the study. However, some males did not ejaculate within 20 min during the first 1-2 times of being placed with an estrous female for testing. Therefore, the first test trial in which any one male ejaculated was recorded as Trial 1 for that male rat. A Mann-Whitney U comparison showed no difference ($p > .05$) in the number of extra trials required across all groups.

Statistical Analysis

Statistical analyses were performed on the latency times to mount, intromit, and ejaculate by using a 3 x 8 (Groups x Trials) mixed design analysis of variance (ANOVA). Individual trials were compared using one-way ANOVA's. Post-hoc *t* tests were conducted on significant results; two-tailed tests of significance were used for all comparisons. If a male failed to mount, intromit, or ejaculate within the time limits (10, 15, 20 min respectively), a score of 600 s was assigned for the mount response, 900 s for the intromission response, and 1200 s for ejaculation.

Results

The scores of the control groups (sex/control, illness/control, and odor/control) were combined because no significant differences were found across the three groups in latency to mount, $F(2,12) < 1$, $p > .05$, intromit, $F(2,12) < 1$, $p > .05$, or ejaculate, $F(2,12) < 1$, $p > .05$. Data were transformed into logarithms (base 10). The raw scores and the standard errors of the transformed data for each copulatory response are given in Appendix B. One male rat died during the experiment and its data were omitted. Results were analyzed for groups: (a) sex/ill (n=9), (b) odor-sex/ill (n=10), and (c) control (n=15).

The mean percentage of ejaculation responses by each group for each trial is depicted in Figure 3. Males in the odor-sex/ill group exhibited ejaculation responses significantly less often than the sex/ill or control groups, $F(2,31) = 20.33$, $p < .0001$. The median trial during which males in the odor-sex/ill group first failed to ejaculate was Trial 4 (range 2-6). In contrast, all males in the sex/ill group exhibited an ejaculatory response through Trial 6, and only three males failed to ejaculate during the last two trials.

Mean latencies for each group for mount, intromission, and ejaculation responses are depicted in Figure 4. Significant group effects were found in latency to mount, $F(2,31) = 80.93$, $p < .0001$, intromit, $F(2,31) = 88.42$, $p < .0001$, and

Figure Caption

Figure 3. Mean percentage of male rats that exhibited ejaculatory responses in Experiment 1.

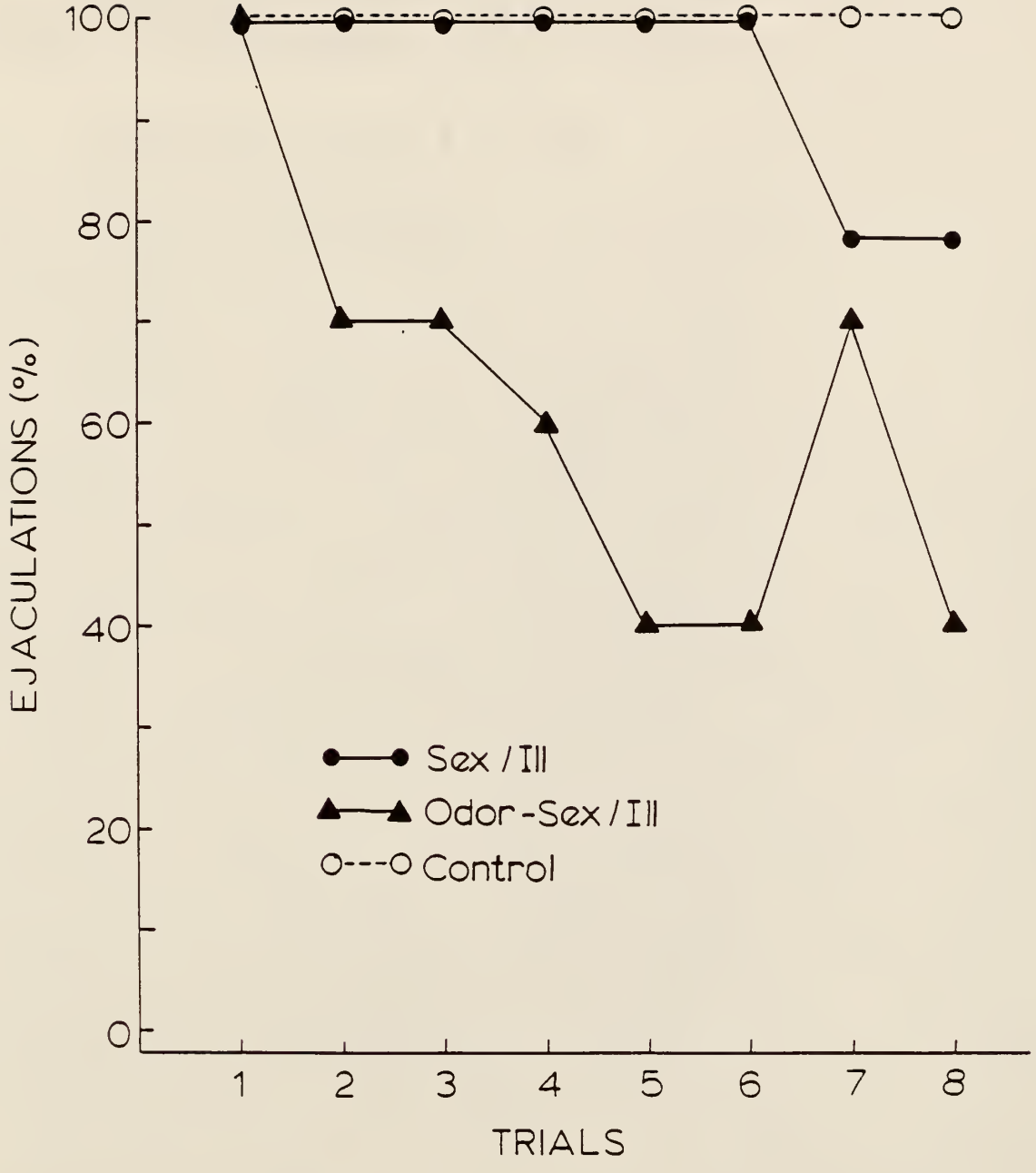
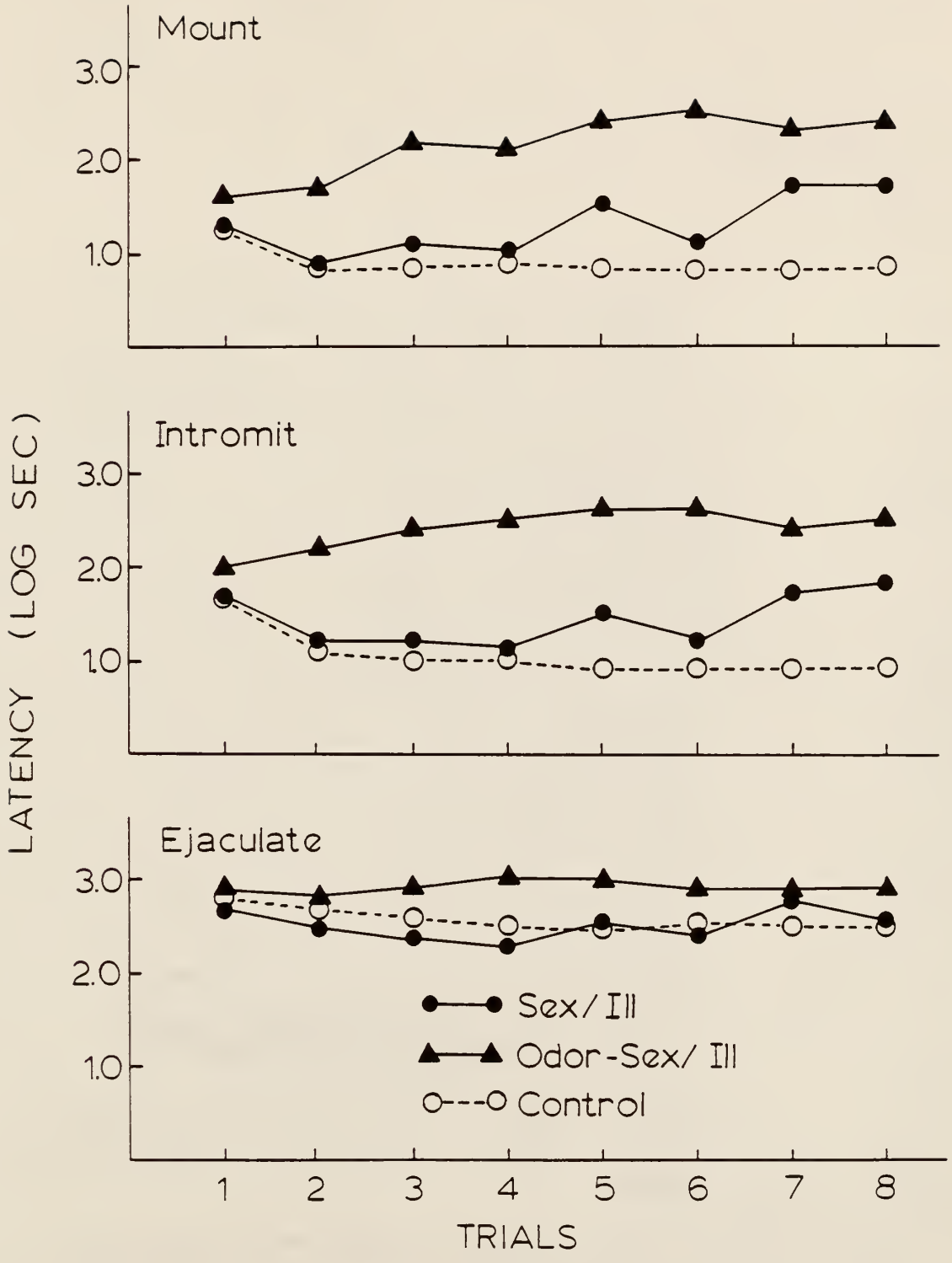

100
80
60
40
20
0
EJACULATIONS (%)
1
2
3
4
5
6
7
8
TRIALS
Sex / Ill
Odor-Sex / Ill
Control

Figure Caption

Figure 4. Mean latency times for mount, intromission, and ejaculation responses in Experiment 1.


Mount
Intromit
Ejaculate
LATENCY (LOG SEC)
3.0
2.0
1.0
1
2
3
4
5
6
7
8
TRIALS
Sex/ Ill
Odor-Sex/ Ill
Control

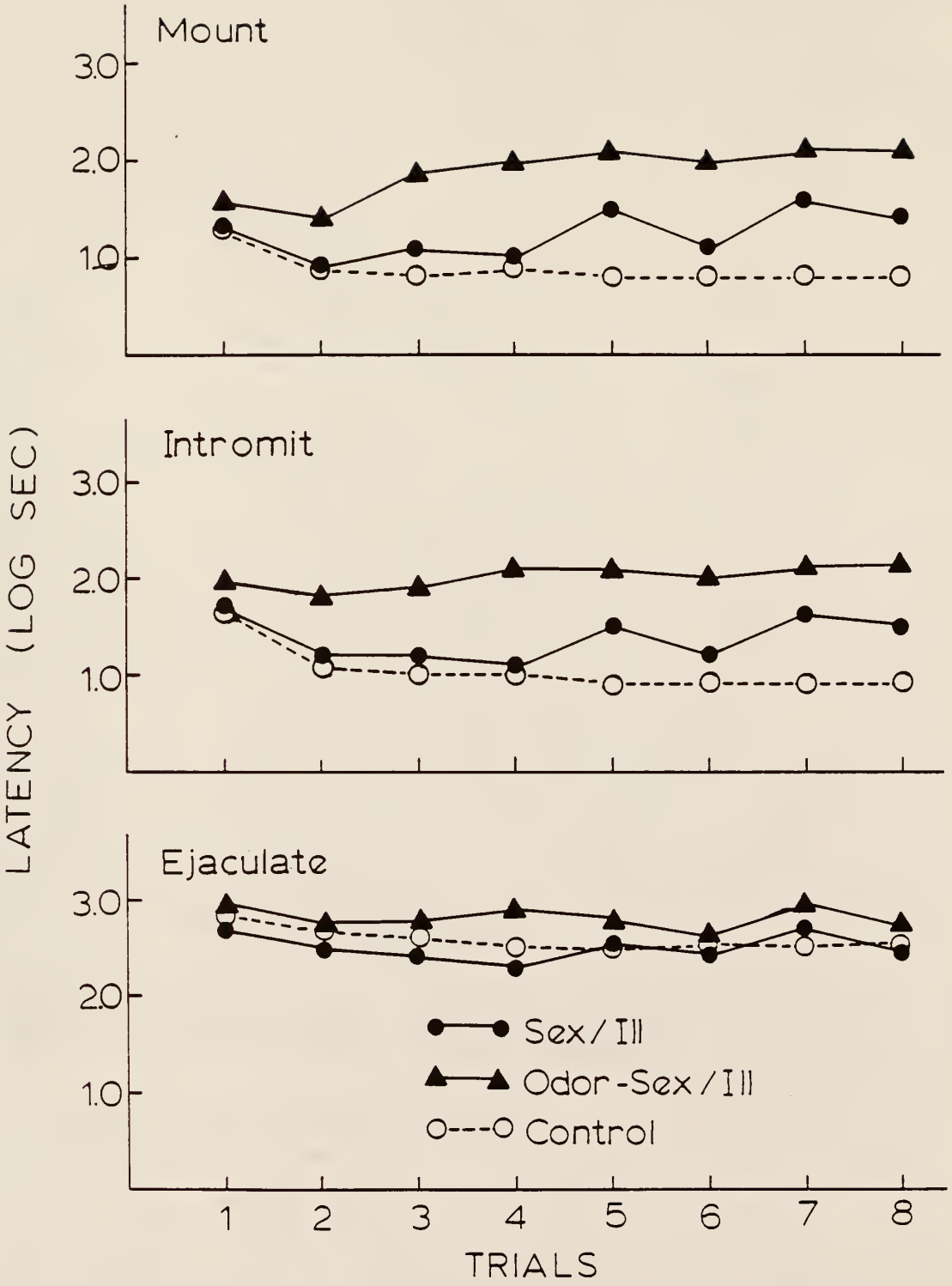
ejaculate, $F(2,31) = 40.27$, $p < .0001$. Post-hoc t tests indicated all three groups differed significantly ($p < .001$) from each other in latency to mount and intromit; the odor-sex/ill group differed significantly from the sex/ill and control groups in latency to ejaculate ($p < .0001$). No significant difference was observed between Trials 8 and 9 in the latency to mount, intromit, or ejaculate by either the odor-sex/ill group or the odor/control group.

An additional analysis was performed on the latencies with a more conservative method of assigning latency times for non-performance. If a male rat failed to mount within 10 min, a score of 600 s was assigned not only to the mount latency but also to the intromission and ejaculation response times. If a male mounted but failed to intromit within 15 min, a score of 900 s was assigned to the intromission and ejaculation times. Males that mounted and intromitted but failed to ejaculate within 20 min were assigned scores of 1200 s for the ejaculation latency. An analysis of the scores obtained by the more conservative method showed the same basic pattern of results as those obtained by the less conservative method.

Additional analyses were performed on the latency times for mount, intromission, and ejaculation responses of each trial by using the latency scores recorded only for the males that exhibited ejaculation responses. The mean latencies for each group are depicted in Figure 5 for mount, intro-

Figure Caption

Figure 5. Mean latency times for mount, intromission and ejaculation responses for males that showed an ejaculation response in Experiment 1.


Mount
Intromit
Ejaculate
LATENCY (LOG SEC)
3.0
2.0
1.0
1
2
3
4
5
6
7
8
TRIALS
Sex / Ill
Odor-Sex / Ill
Control

mission, and ejaculation responses. Because males that failed to ejaculate were omitted from this analysis, the number of rats per group per trial varied, especially in the odor-sex/ill group. The sample size of each group for each trial is presented in Table 2. The odor-sex/ill group differed significantly ($p < .05$) from the control and sex/ill groups for mount and intromission responses on all trials except Trial 1. The odor-sex/ill group differed significantly ($p < .05$) in latency to ejaculate from the sex/ill group (Trials 2, 3, 4, 5, 8) and from the control group (Trials 3, 4, 5, 7, 8). The sex/ill group differed significantly ($p < .05$) from the control group for latency to mount (Trials 3, 5, 7, 8), intromit (Trials 5, 7, 8), and ejaculate (Trials 2, 3, 4, 7).

The mean number of intromissions to ejaculation by males that exhibited an ejaculation response are depicted in Figure 6. No significant difference was noted ($p > .05$) between the odor-sex/ill and sex/ill groups. The control group showed significantly more ($p < .05$) intromissions than the sex/ill and odor-sex/ill groups in half the trials (Trials 3, 4, 6, 8).

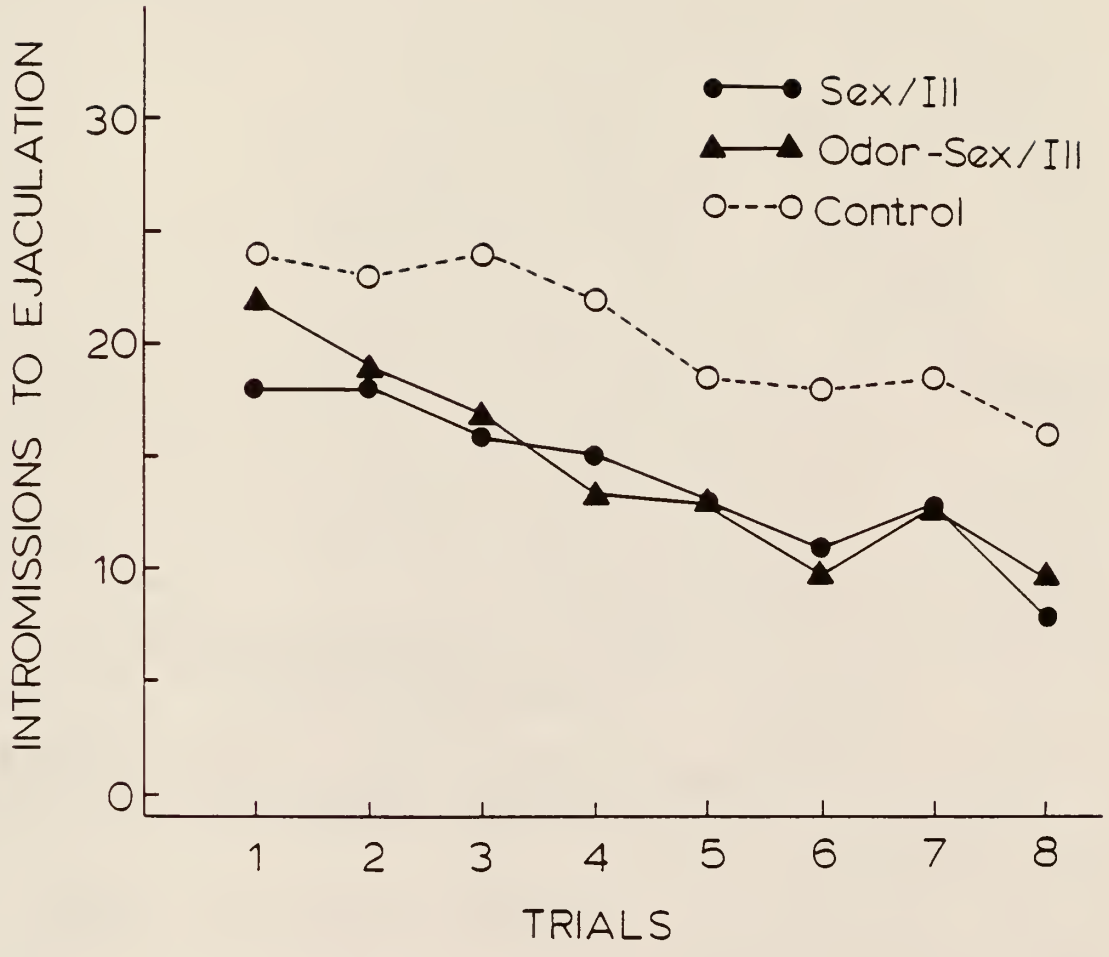
Table 2

Group Sample Sizes of Rats Exhibiting Ejaculatory Responses in each Trial of Experiment 1

	Trial							
Group	1	2	3	4	5	6	7	8
Sex/Ill	9	9	9	9	9	9	7	7
Control	15	15	15	15	15	15	15	15
Odor-Sex/Ill	10	7	7	6	4	4	7	4

Figure Caption

Figure 6. Mean number of intromissions to ejaculation by males exhibiting an ejaculation response in Experiment 1.


INTROMISSIONS TO EJACULATION
30
20
10
0
1
2
3
4
5
6
7
8
TRIALS
Sex/Ill
Odor-Sex/Ill
Control

Discussion

Males in the odor-sex/ill group showed a significant difference in latency to mount, intromit, and ejaculate when compared with the sex/ill and control groups. Also, males in the odor-sex/ill group exhibited significantly fewer ejaculation responses than the sex/ill or control groups. By Trial 6 every male in the odor-sex/ill group had failed to exhibit an ejaculatory response at least one time; two males in the odor-sex/ill group failed to exhibit an ejaculatory response during five consecutive trials. It is suggested the odor served as a potentiating cue to produce a copulatory aversion. Because no significant difference was found in latencies across all groups in Trial 1, it is suggested the aversion noted in the odor-sex/ill group was not due to a neophobic or aversive response to a novel odor. The non-aversiveness of the novel odor is further supported by the fact that rats in the odor/control group showed normal copulatory behavior throughout testing.

Males in the odor-sex/ill and odor/control groups were tested an additional trial (Trial 9) in which a non-odorous, estrous female was placed into the testing chamber with the male. No significant difference was observed for latencies to mount, intromit, or ejaculate between Trials 8 and 9 by the odor-sex/ill or odor/control groups.

Comparing the data obtained only from males that showed an ejaculation response, longer latencies to mount, intromit,

and ejaculate were observed in the odor-sex/ill group. Also, the number of intromissions to ejaculation was significantly fewer than the control group on most trials.

The results of Experiment 1 showed males made ill following ejaculation without the odor cue failed to show a significant aversion to copulatory behavior; these results do not support those found by Peters (1983). Only two of the nine males in the sex/ill group failed to ejaculate during one trial (Trials 7 or 8), but did ejaculate during the following trial. One additional male in the sex/ill group failed to ejaculate during two consecutive trials (Trials 7 and 8). The two males that did not ejaculate during Trial 8 were observed an extra trial (Trial 9) to test extinction of the aversion; both males ejaculated during Trial 9. As in the Pilot Study, results suggest the aversion extinguished after one trial in which LiCl was not given in the sex/ill group.

Peters (1983) reported an aversion to copulatory behavior was characterized by the males' showing neither approach nor investigation of the female. Males in the sex/ill group in Experiment 1 showed significantly longer latencies to mount and intromit than the control males, but only in Trial 8 did two males fail to exhibit mount and intromission responses. These results do not support those reported by Peters.

EXPERIMENT 2

Results of the Pilot Study and Experiment 1 indicated an aversion to copulatory behavior by male rats was potentiated by a novel odor. Peters (1983) reported male rats ceased mating behavior after LiCl illness following each 30 min encounter with an estrous female. The purpose of Experiment 2 was to replicate the copulatory aversion reported by Peters with non-contingent LiCl-induced illness. Intragastric LiCl was used in Experiment 2, rather than injections of LiCl used by Peters. The same odor-sex/ill group described in the Pilot Study and Experiment 1 was observed in Experiment 2 to test whether a novel odor cue potentiated a non-contingent aversion to copulatory behavior.

Method

Subjects (n=40), apparatus, and procedure specifics were the same as those described in Experiment 1, except intubations were given all males after ejaculation (as described previously) or at the end of a 20 min encounter with an estrous female, independent of the mating responses exhibited during the test trials. All males were left in the testing chamber 20 min unless ejaculation had occurred previous to that time. Latency scores were recorded for mount, intromission, and ejaculation responses; a score of 1200 s was assigned to each copulatory response not exhibited by the males. The number of intromissions to ejaculation was recorded for males that exhibited an ejaculatory response. Extinction trials were conducted, and the trial of first ejaculation was recorded. The extinction trials were conducted exactly the same as the test trials, except intubations were not performed. The odor-sex/ill and odor/control groups were presented an odorous female.

Statistical Analysis

The same analyses were performed as described in Experiment 1, except times of 1200 s were assigned for non performance of each behavioral response. A one-way ANOVA was performed on the scores of the extinction trials. A Mann-Whitney U comparison showed no difference ($p > .05$) in the number of trials required across all groups to attain the criterion of Trial 1.

Results

The scores of the three control groups (sex/control, illness/control, and odor/control) were combined for the analyses because no significant differences were noted across the three groups in latency to mount, $F(2,12) = 1.2$, $p > .05$, intromit, $F(2,12) < 1$, $p > .05$, or ejaculate, $F(2,12) < 1$, $p > .05$. The data were transformed into logarithms (base 10); the raw scores and the standard errors for the transformed data are presented in Appendix C. One male rat died during the experiment and its data were omitted. One additional male in the sex/ill group died after Trial 8; its data were included in the analyses through Trial 8. Results were analyzed for groups: (a) sex/ill (n=10 except during extinction trials where n=9), (b) control (n=15), and (c) odor-sex/ill (n=9).

The mean percentage of ejaculation responses by each group for each trial is depicted in Figure 7. Males in the odor-sex/ill group exhibited ejaculation responses significantly less often than the sex/ill or control groups, $F(2,31) = 29.47$, $p < .0001$. The mean and median trial in which males in the odor-sex/ill group failed to ejaculate was Trial 4 (range 2-8). In contrast, only three males in the sex/ill group failed to exhibit an ejaculatory response throughout the eight trials (Trials 4, 7, 7).

Mean latencies for each group for mount, intromission, and ejaculation responses are depicted in Figure 8. Signifi-

Figure Caption

Figure 7. Mean percentage of male rats that exhibited ejaculatory responses in Experiment 2.

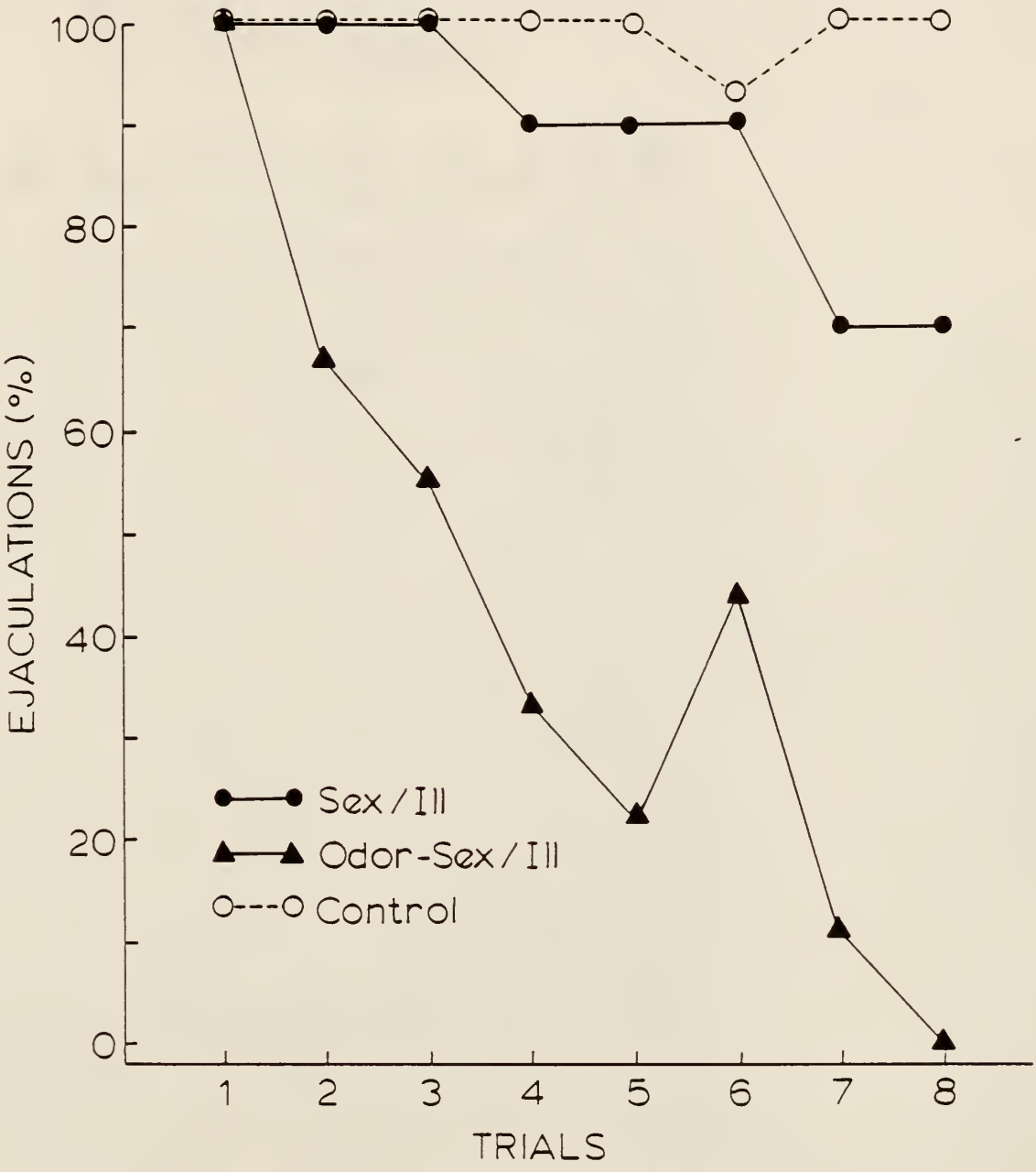
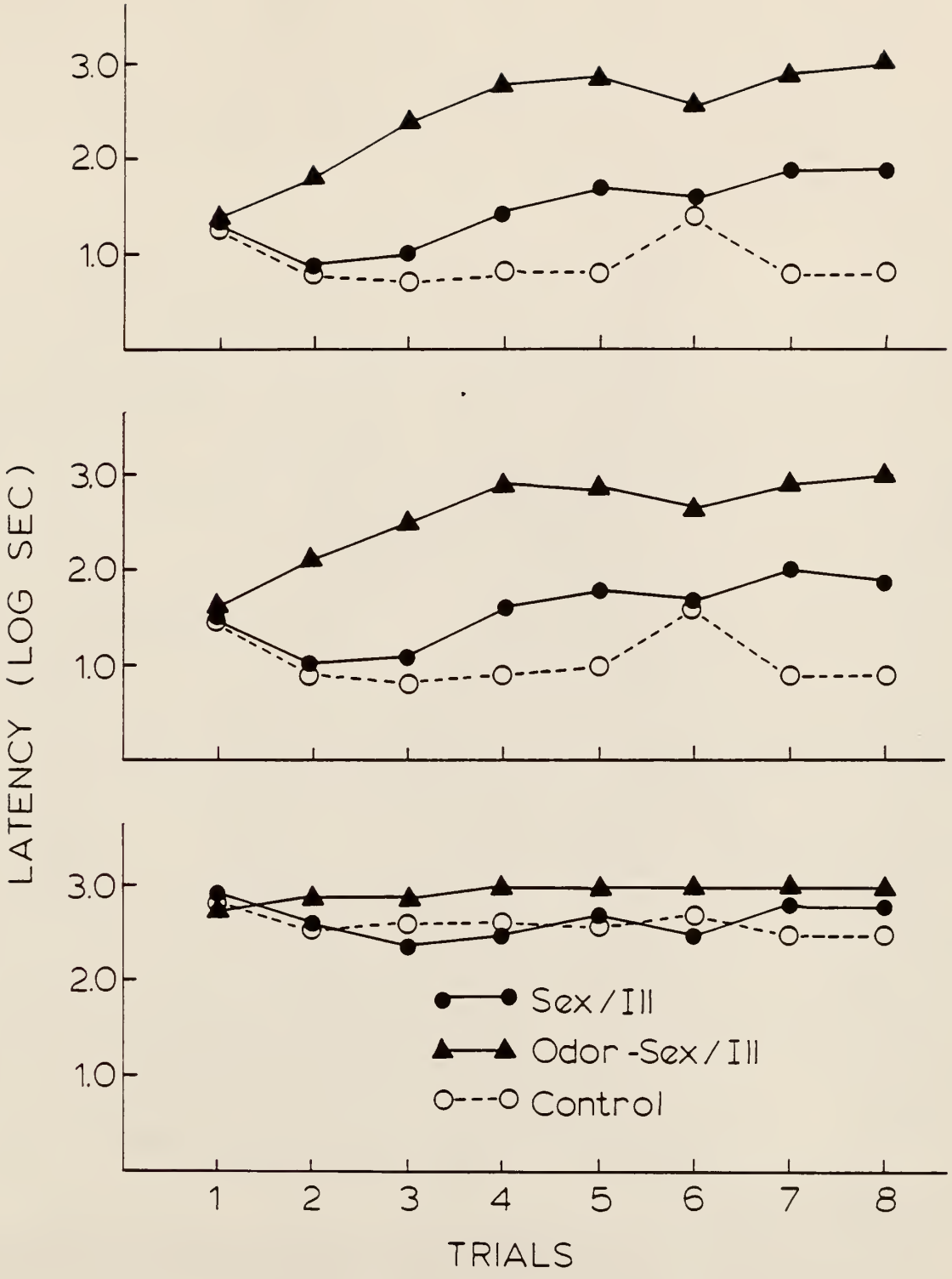

100
80
60
40
20
0
EJACULATIONS (%)
1
2
3
4
5
6
7
8
TRIALS
Sex / Ill
Odor-Sex / Ill
Control

Figure Caption

Figure 8. Mean latency times for mount, intromission, and ejaculation responses in Experiment 2.


LATENCY (LOG SEC)
3.0
2.0
1.0
3.0
2.0
1.0
3.0
2.0
1.0
Sex / Ill
Odor -Sex / Ill
Control
1
2
3
4
5
6
7
8
TRIALS

cant group effects were found in latency to mount, $F(2,31) = 100.15$, $p < .0001$, intromit, $F(2,31) = 76.08$, $p < .0001$, and ejaculate, $F(2,31) = 49.27$, $p < .0001$. Post-hoc t tests indicated all three groups differed significantly ($p < .001$) from each other in latency to mount and intromit; the odor-sex/ill group differed significantly ($p < .001$) from the sex/ill and control groups in latency to ejaculate. An additional analysis was performed on the latencies of mount, intromission, and ejaculation responses of each trial by using the latency scores recorded only for the males that exhibited ejaculation responses. The mean latencies for each group are depicted in Figure 9. Because males that failed to ejaculate were omitted from this analysis, the number of rats per group per trial varied, especially in the odor-sex/ill group. The sample size of each group for each trial is presented in Table 3. Data from males in the odor-sex/ill group were not included in the analyses of Trials 7 and 8 because the sample size was 1 and 0 for these two trials.

The odor-sex/ill group differed significantly from the sex/ill and control groups ($p < .05$) for latency to mount on all analyzed trials except Trial 1. The sex/ill group differed significantly ($p < .05$) from the controls for mount latencies on half the trials (Trial 4, 5, 7, 8). The odor-sex/ill group differed significantly ($p < .05$) from the controls for latency to intromit on all analyzed trials except Trial 1, and differed significantly ($p < .05$) from

Figure Caption

Figure 9. Mean latency times for mount, intromission, and ejaculation responses for males that showed an ejaculation response in Experiment 2. Note that no data point is depicted for the odor-sex/ill group for Trial 8, as none in the group ejaculated.

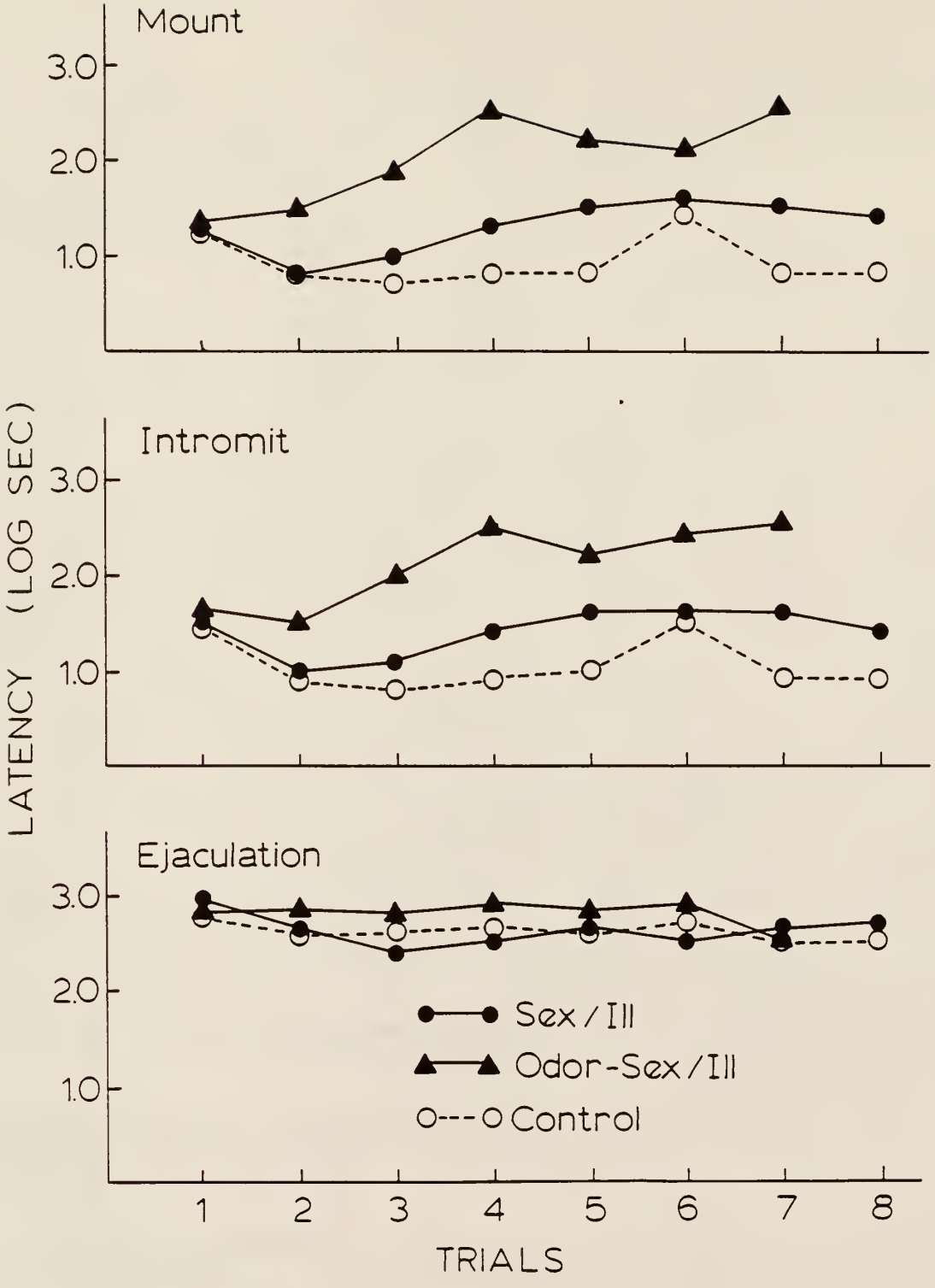

Mount
Intromit
Ejaculation
LATENCY (LOG SEC)
3.0
2.0
1.0
Sex / Ill
Odor-Sex / Ill
Control
1
2
3
4
5
6
7
8
TRIALS

Table 3

Group Sample Sizes of Rats Exhibiting Ejaculatory Responses in each Trial of Experiment 2

	Trial							
Group	1	2	3	4	5	6	7	8
Sex/Ill	10	10	10	9	9	9	7	7
Control	15	15	15	15	15	14	15	15
Odor-Sex/Ill	9	6	5	3	2	4	1	0

the sex/ill group on all analyzed trials except Trials 1 and 5. The sex/ill group differed significantly ($p < .05$) from the controls in latency to intromit on Trials 3, 4, 5, 7, and 8.

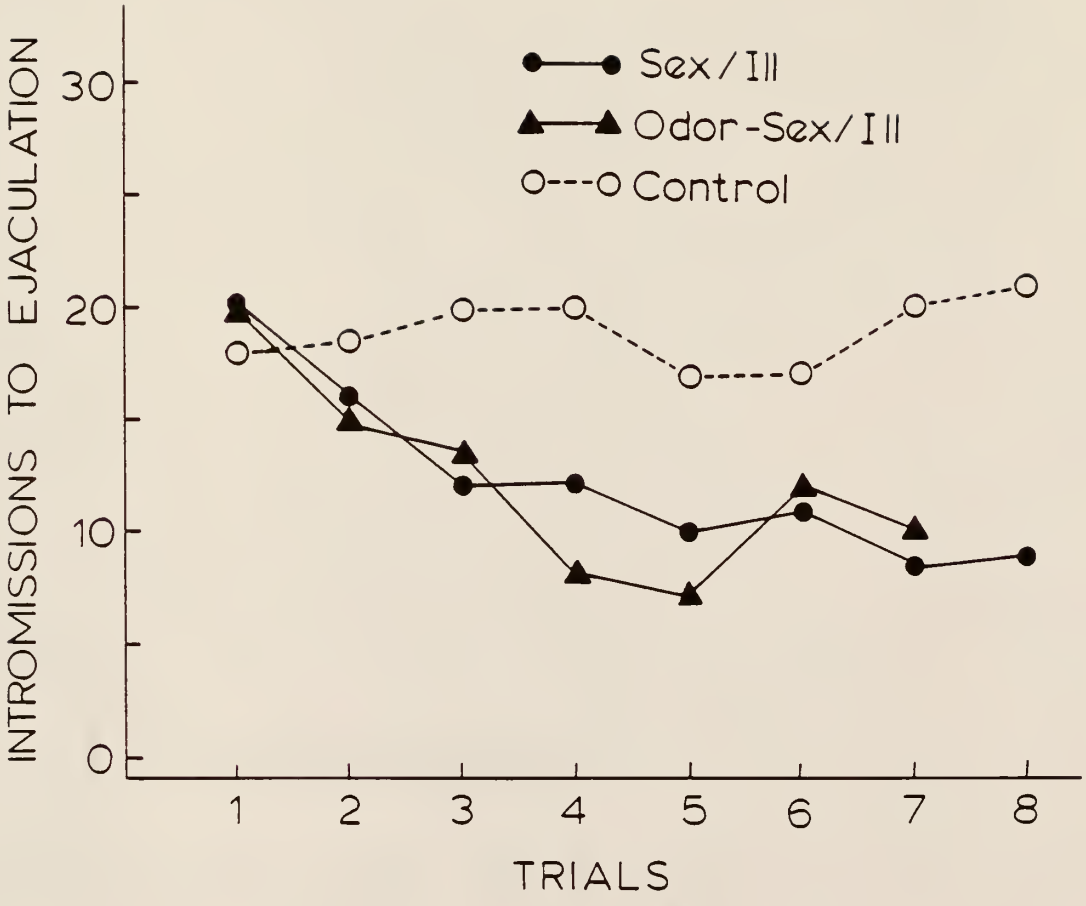
The odor-sex/ill group differed significantly ($p < .05$) from the sex/ill and control groups in latency to ejaculate on all analyzed trials except Trials 1 and 5. The sex/ill group differed significantly ($p < .05$) from the controls in latency to ejaculate on Trials 6, 7, and 8.

The mean number of intromissions to ejaculation by the males that exhibited an ejaculation response are depicted in Figure 10. The controls showed significantly more intromissions to ejaculation ($p < .05$) than the two experimental groups on all trials after Trial 3.

Mean extinction trials in which ejaculations were first observed were 1.0 for the controls, 1.4 for the sex/ill group, and 4.0 for the odor-sex/ill group. The analysis indicated males in the odor-sex/ill group required significantly more trials, $F(2,30) = 109.85$, $p < .0001$, to extinguish the aversion than the males in the sex/ill group. Two males in the odor-sex/ill group ejaculated during Extinction Trial 3, five males in Trial 4, and two males in Trial 5. All males in the control group, of course, ejaculated on the first extinction trial.

Figure Caption

Figure 10. Mean number of intromissions to ejaculation by males exhibiting an ejaculation response in Experiment 2. Note that no data point is depicted for the odor-sex/ill group for Trial 8, as none in the group ejaculated.


Sex / III
Odor-Sex / III
Control
INTROMISSIONS TO EJACULATION
30
20
10
0
1
2
3
4
5
6
7
8
TRIALS

Discussion

Results from Experiment 2 replicated those of the Pilot Study and Experiment 1; the novel odor potentiated a copulatory aversion. Compared with the sex/ill and control groups, the odor-sex/ill group exhibited significantly longer latencies to mount, intromit, and ejaculate. Males in the odor-sex/ill group exhibited significantly fewer ejaculation responses; less than 100% ejaculation responses were observed in every trial after Trial 1. The odor-sex/ill group required significantly more non-reinforced trials to extinguish the copulatory aversion. It was concluded that a strong aversion was formed by the males in the odor-sex/ill group, an aversion significantly stronger than that observed in the males in the sex/ill group. As in Experiment 1, no significant difference was noted in latencies across all groups in Trial 1 of Experiment 2, and males in the odor/control group formed no aversion to copulatory behavior; thus, it was concluded the aversion was not due to an aversive response to a novel odor.

Males in the sex/ill group that were made ill following a 20 min encounter with an estrous female and/or after an ejaculatory response, without the odor cue, failed to show a significant aversion to copulatory behavior. Compared with the controls, the sex/ill group failed to show a significant difference in the percentage of ejaculation responses. Significantly longer latencies to ejaculate were noted in

the sex/ill group during the last three trials; two males in the sex/ill group failed to exhibit an ejaculatory response during the last two trials, and one male failed to do so on five consecutive trials. No significance was noted between the sex/ill and control groups in the number of non-reinforced trials to extinction.

GENERAL DISCUSSION

A novel odor significantly potentiated a learned aversion to copulatory behavior in male rats in three experiments of the present study. The potentiation was evidenced by significant changes in: (a) longer latency times to mount, intromit, and ejaculate, (b) a lower percentage of males that exhibited an ejaculatory response, and (c) increased resistance to extinction of the aversion. Although not empirically tested, males in the odor-sex/ill group also displayed behavior changes other than changes in mating responses.

Peters (1983) reported a copulatory aversion in male rats made ill with LiCl injections, non-contingent on the copulatory behavior displayed. Although 9 of 23 male rats in the sex/ill group of the present study failed to ejaculate in 17 trials (11.5% of the total trials), the aversion was not so strong as that reported by Peters. No difference was noted in the percentage of ejaculation responses displayed by the sex/ill group whether the LiCl-induced illness was contingent (Experiment 1) or non-contingent (Experiment 2) on copulatory responses. Because males in the sex/ill group ejaculated on most trials in Experiment 1, and thus were made ill, the treatment conditions for this group were virtually identical in Experiments 1 and 2.

One male in the sex/ill group during Experiment 2 failed to ejaculate on five consecutive trials, but no other male

in the sex/ill group displayed an aversion for more than two of eight trials. The aversion noted in the sex/ill group extinguished after one non-reinforced trial, except that the one male that failed to ejaculate on five consecutive trials required two non-reinforced trials before it displayed an ejaculatory response. No significant difference was found in the number of extinction trials between the sex/ill and control groups.

It is suggested the sex/ill group in the present study failed to replicate the significant aversion reported by Peters (1983) as a function of the different method of administering LiCl. LiCl was administered intragastrically by intubation in the present study, but by intraperitoneal injection by Peters. It is suggested the painful stimulus of the injections in Peters' study facilitated the illness stimulus to produce a copulatory aversion. Lasiter and Braun (1981) have reported a similar type of facilitative effect of weak punishment cues in taste aversion learning. Shock alone failed to produce a taste aversion, but shock administered just prior to rotation-induced illness facilitated the aversion. Lacking the facilitative pain stimulus in the present study, a significant aversion was not noted in the sex/ill group.

Combined stimuli (illness and pain) for the learned copulatory aversion in Peters' (1983) study is likened to the potentiating effect of the novel odor in the present

study. It has been shown (Rusiniak et al., 1979) that odor is a weak cue in taste aversion learning, but that the odor cue was potentiated (facilitated) when paired with the strong cue of taste. While offering an explanation of taste aversion learning, Garcia and Koelling (1966) suggested certain stimuli are selectively associated with certain visceral events and behaviors. Because odor stimuli are intimately associated with sexual behavior in the rat (Beach, 1956; Beach, 1976; Caroom & Bronson, 1971; Noble, 1973; Pfaff et al., 1973; Powers & Winans, 1975), it is a logical conclusion to believe modification of odor stimuli would be a strong cue for modifying sexual behavior. This conclusion supports the belongingness (Seligman, 1970) notion that particular cues are associated with specific consequences.

Modification of copulation (an aversion) is of particular interest because it is non-adaptive behavior not to mate. A taste aversion to avoid harmful substances is sound, adaptive behavior, but a complete copulatory aversion would result in extinction of the species. Males in the odor-sex/ill group required significantly more trials to extinguish the copulatory aversion than did the sex/ill and control groups; however, the extinction was accomplished more rapidly than expected. Although two of nine males in the odor-sex/ill group in Experiment 2 showed no copulatory responses after four non-reinforced trials, seven of nine extinguished by the fourth trial. Perhaps the aversion extinguished

relatively quickly because of the non-adaptive aspect of the aversion.

When present, the copulatory aversion was characterized by cessation of all responses associated with copulatory behavior, including approach and investigation of the female (Beach, 1956). Males in the odor-sex/ill group portrayed "submissive" behavior by crouching in the corner of the testing chamber and showing neither aggression nor self-defense behaviors even when the estrous female bit the male. One male in the odor-sex/ill group shook violently while crouched in the corner; another male repeatedly jumped out of the testing chamber, even when presented a non-odorous female.

The nine males in the sex/ill group that failed to ejaculate displayed none of the same behaviors observed in the males in the odor-sex/ill group. Males in the sex/ill group sat in the corner of the testing chamber grooming, or chased the estrous female after the female had approached the male. When the female exhibited the lordosis posture during the chase, on some occasions the males approached from the rear and crawled over the female. Some chases were accompanied by "pseudointromission" responses; the males palpated the female and sprang off, but showed no auto-genital cleaning behavior. After one or two such responses, the males would return to the corner of the testing chamber. At other times the males only chased the female, then

returned to the corner.

Observation of some behaviors displayed by the males in the odor-sex/ill group in the present study agreed with Peters (1983) who reported that one male learned to escape from the testing chamber, and none displayed overt aggressive behavior when attacked. However, observations were not noted to agree with Peters who reported males kicked at the females and displayed paw-treading movements of the floor and sides of the chamber. Some males in the odor-sex/ill group did stand on hind feet while pawing the sides of the testing chamber with their fore feet, but this behavior was attributed to escape attempts rather than actual paw-treading movements of "agitation" and "disgust" reported by Peters. Paw-treading the floor of the chamber and chin-rubbing behaviors were not observed in the present study, in contrast to the observations of Peters. Because no attempt to escape was noted during the midinterval tests, the escape maneuvers were believed to be specific to the odorous female and not to the testing chamber.

An interesting result of the present study was the significantly reduced number of intromissions to ejaculation shown by the two experimental groups relative to the controls. Beach and Fowler (1959) reported a "situational anxiety" state in which males ejaculated after fewer intromissions than typically observed. When a male was aroused by stress of previous shock treatments, fewer intromissions were needed

to summate to an ejaculatory threshold for an ejaculation response. Although significantly more intromissions to ejaculation were observed in the controls than in the two experimental groups, latency times for mount and intromission responses were more prolonged in the experimental groups. Males in the sex/ill group showed longer latencies than control males to mount and intromit, but the significantly fewer intromissions to ejaculation resulted in equal or slightly less latencies to ejaculate in the sex/ill group compared with controls.

As stated by Beach (1956), males that failed to exhibit a mount response within 10 min were unlikely to do so, even if tested for one hour. Data from the present study support this conclusion; if mount responses were observed during the 20 min test trial, they occurred within the first 10 min. This observation supports the validity of the scores (600 s for mount, 900 s for intromission, 1200 s for ejaculation) assigned to non-performance latencies in the present study. Indeed, the assigned values may even be underestimates of the actual values, assuming the male rats were allowed longer test periods.

Previous studies in hamsters (Emmerick & Snowdon, 1976; Johnston et al., 1978) suggested mating behavior was not affected by LiCl poisoning unless the poison was paired with ingestion of vaginal secretions to produce an aversion. Results were reported of increased latency times to mount

only; intromission and ejaculation responses were not altered. Males in the present study that were made ill following ejaculation (odor-sex/ill and sex/ill groups) showed increased latencies for mount responses also; however, males in the present study also exhibited increased latencies for intromission and ejaculation responses. Furthermore, complete cessation of copulatory responses were observed, especially in the odor-sex/ill group.

An increased percentage of males exhibited copulatory behavior in Experiments 1 and 2, compared with the Pilot Study, before illness was induced. The improved number of sexually active males is attributed to housing the males five per cage upon arrival to the laboratory (Peters & Blythe, 1983), and testing the males during the dark phase of the diurnal cycle (Harlan et al., 1979).

Data from the present study suggest the observed aversion was to copulatory behavior and not to odor. The failure of males in the odor-sex/ill group to display copulatory behavior when presented a non-odorous female in Experiment 1 supports the interpretation of a copulatory aversion. Regardless of whether the aversion was to copulatory behavior or to a novel odor, modification of mating behavior was accomplished. According to the Garcia and Rusiniak (1980) indexing hypothesis, an odor cue present during copulation would be indexed in a reproductive or copulatory context and not in another context (feeding

behavior). If, however, males showed an aversion to the odor in situations other than mating situations (e.g. a consummatory context), one would conclude the odor had assumed more general aversive properties, ones not tied to any specific behavior.

Because the olfactory system, and the vomeronasal system in particular, have been shown to influence mating behavior (Powers & Winans, 1975), further support for the interpretation of a copulatory aversion may be obtained by modifying the olfactory and vomeronasal systems. A future study may produce a copulatory aversion with a novel odor cue in male rats, then destroy the olfactory and vomeronasal systems; a persistent aversion would support the interpretation of a copulatory aversion, because no odor stimuli could be detected.

Results of the present study suggest that mating behavior is not strictly an automatic response to changing hormone levels; modifications of copulatory behavior have been shown to be a process of learning. Although olfaction has long been accepted to play a role in mating behavior, very little research has been conducted to study the associations between odor cues and mating behavior. Results of the present study have shown a basic behavior, one necessary for the preservation of the species, was modified by utilizing only a neutral odor paired with illness.

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Appendices

Note: * signifies no responses were noted.

Appendix A-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in the Pilot Study--odor-sex/ill group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	70	15	90	185	*	195	*	85
4	360	105	95	185	*	195	*	85
	915	505	380	510	*	440	*	500
	120	140	270	95	*	140	60	*
16	190	*	300	95	*	140	60	*
	895	*	1190	420	*	600	520	*
	320	55	55	*	*	*	5	155
21	320	100	825	*	*	*	145	*
	1180	*	*	*	*	*	1030	*
	20	55	310	45	105	200	170	*
26	20	55	310	45	105	200	170	*
	465	510	*	425	320	430	430	*

Appendix A-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in the Pilot Study--sex/ill group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	35	90	245	60	*	305	230	90
6	120	105	245	110	*	340	250	105
	685	785	565	675	*	700	890	410
	200	60	70	30	155	175	150	205
7	795	65	70	75	155	175	*	205
	1195	465	370	380	405	*	*	440
	100	70	130	125	105	115	65	110
11	145	85	230	135	110	115	65	115
	1065	575	725	1095	690	485	1165	685
	65	5	65	55	65	115	255	95
30	80	50	105	55	65	115	255	95
	580	420	980	430	*	760	1195	645

Appendix A-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in the Pilot Study--illness/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	10	5	10	65	25	15	45
1	40	80	35	10	105	45	120	45
	625	*	615	375	440	340	505	435
	10	5	20	10	10	5	65	10
8	10	5	20	10	10	15	90	10
	255	405	255	275	205	275	340	270
	35	10	10	35	5	25	10	35
13	85	15	10	35	5	25	10	90
	765	595	355	385	595	465	525	495
	5	45	35	45	130	195	130	70
24	5	45	45	95	130	195	135	70
	790	455	410	530	335	715	465	455

Appendix A-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in the Pilot Study--sex/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	75	15	10	30	5	40	5	140
12	90	15	15	30	10	40	5	140
	330	390	235	450	625	420	305	330
	50	55	80	195	185	130	135	*
18	180	285	*	*	660	180	140	*
	680	*	*	*	*	705	675	*
	90	20	10	85	135	250	140	95
19	150	35	85	105	170	355	140	125
	625	220	405	425	425	705	*	435

Appendix A-2

Standard errors of the log transformed data of latencies for mount, intromission, and ejaculation responses in the Pilot Study, Figure 2.

Trial	Mount	Intromission	Ejaculate
	Odor-Sex/Ill Group		
1	0.2174	0.2535	0.0747
2	0.1726	0.2305	0.0934
3	0.1585	0.1662	0.1079
4	0.2067	0.2399	0.0938
5	0.1639	0.2020	0.1243
6	0.1193	0.1564	0.0900
7	0.3820	0.2126	0.0947
8	0.1854	0.2219	0.0823
	Sex/Ill Group		
1	0.1379	0.1904	0.0651
2	0.2531	0.0606	0.0520
3	0.1165	0.1150	0.0774
4	0.1100	0.0752	0.0905
5	0.1796	0.2142	0.0980
6	0.0869	0.0963	0.0700
7	0.1169	0.2018	0.0272
8	0.0712	0.0649	0.0491
	Control		
1	0.1723	0.2091	0.0670
2	0.1305	0.1995	0.0943
3	0.1424	0.2285	0.0847
4	0.1636	0.2393	0.0703
5	0.2469	0.2781	0.0839
6	0.2119	0.1811	0.0589
7	0.2137	0.2154	0.0693
8	0.1927	0.2055	0.0714

Appendix B-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 1--odor-sex/ill group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	35	10	40	75	*	10	15	115
10	95	10	50	75	*	10	15	120
	300	270	310	890	*	120	735	235
	110	10	95	240	350	*	135	75
11	350	215	150	240	350	*	135	75
	1080	840	985	835	1150	*	830	*
	20	30	70	260	*	120	190	180
14	90	105	95	260	*	120	190	190
	870	680	400	635	*	260	520	400
	5	40	*	*	135	*	120	*
17	50	*	*	*	*	*	310	*
	765	*	*	*	*	*	1185	*
	60	75	75	190	*	*	180	240
18	215	120	135	190	*	*	180	240
	890	550	515	1050	*	*	540	1090
	85	45	*	60	15	460	*	*
21	90	60	*	70	25	685	*	*
	515	180	*	330	280	970	*	*
	5	*	*	120	*	*	*	*
22	10	*	*	120	*	*	*	*
	765	*	*	995	*	*	*	*
	15	*	250	*	*	*	*	*
23	60	*	270	*	*	*	*	*
	790	*	1030	*	*	*	*	*
	385	20	240	425	115	*	390	*
28	385	20	350	*	115	*	390	*
	1005	470	705	*	500	*	960	*
	140	35	15	*	295	155	90	60
32	330	190	120	*	295	155	120	60
	860	460	390	*	1080	870	630	600

Appendix B-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 1--sex/ill group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	10	30	15	10	20	70	10
1	20	15	35	20	15	20	70	115
	495	380	310	325	200	170	335	155
	60	30	15	5	75	5	15	65
3	130	30	20	15	75	5	15	65
	910	420	270	260	240	230	245	225
	10	5	10	15	10	25	50	30
4	45	10	10	15	20	25	50	65
	850	690	600	325	335	895	380	720
	60	5	15	10	80	50	70	*
6	60	5	15	10	90	95	80	*
	320	220	130	65	355	330	415	*
	30	5	10	10	10	10	120	80
15	335	10	15	15	10	10	150	90
	855	255	275	285	190	270	1100	165
	10	10	5	5	110	10	15	25
16	10	10	15	10	110	15	15	25
	315	180	200	210	420	295	940	270
	10	5	10	5	15	10	60	10
30	35	20	10	5	15	10	90	10
	305	185	175	90	315	150	*	185
	15	10	15	10	70	10	150	*
31	15	15	20	10	70	10	150	*
	405	300	310	210	250	280	*	*
	30	5	15	35	20	5	15	10
34	240	30	15	45	30	10	15	10
	790	350	315	405	400	195	720	190

Appendix B-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 1--illness/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	5	5	5	5	5	5	5
7	120	40	15	10	10	5	5	10
	885	645	360	215	445	305	290	245
	20	30	10	10	5	10	10	5
9	50	35	25	10	5	10	10	5
	435	555	490	365	390	330	480	275
	60	5	10	10	5	10	10	5
19	165	5	15	10	5	15	10	10
	510	400	350	250	180	365	210	230
	20	5	10	5	5	5	5	5
29	20	5	10	10	5	5	5	10
	945	480	290	330	470	340	250	320
	15	5	5	10	10	10	5	10
35	15	10	5	10	10	10	5	10
	460	495	310	415	260	250	165	255

Appendix B-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 1--sex/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	35	10	5	5	5	5	5	5
12	50	10	5	5	5	5	5	5
	320	590	255	155	250	345	360	300
	15	15	5	5	5	5	5	5
13	20	20	5	10	10	10	10	5
	860	840	370	545	425	395	430	410
	15	10	5	5	10	10	10	5
20	15	10	10	10	10	15	10	5
	365	360	420	180	310	310	255	360
	10	10	5	10	10	5	5	5
26	590	10	5	10	10	5	5	5
	1140	505	315	495	270	330	435	420
	5	5	5	10	5	5	10	15
33	230	15	15	15	5	10	15	15
	1025	300	540	380	415	420	440	415

Appendix B-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 1--odor/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	5	10	10	10	10	10	10	5
2	5	10	10	10	10	10	10	5
	470	235	250	450	250	345	335	210
	60	5	5	10	5	5	5	10
5	195	10	15	15	10	5	5	10
	480	405	470	315	200	235	260	175
	105	5	10	10	10	10	5	15
24	320	25	15	10	10	10	10	15
	980	735	375	390	360	455	420	555
	5	10	5	10	5	5	5	5
25	40	10	5	10	10	15	10	10
	970	430	630	420	260	250	300	275
	10	5	10	5	5	5	5	5
27	15	5	15	10	10	5	5	5
	605	350	585	255	240	355	420	295

Appendix B-2

Standard errors of the log transformed data of latencies for mount, intromission, and ejaculation responses in Experiment 1, Figure 4.

Trial	Mount	Intromission	Ejaculate
	Odor-Sex/Ill Group		
1	0.1871	0.1499	0.0490
2	0.1836	0.2003	0.0846
3	0.1657	0.1356	0.0693
4	0.1121	0.1372	0.0532
5	0.1556	0.1583	0.0656
6	0.1732	0.1951	0.1053
7	0.1493	0.1635	0.0443
8	0.1228	0.1477	0.0767
	Sex/Ill Group		
1	0.1027	0.1633	0.0649
2	0.0807	0.0663	0.0592
3	0.0607	0.0522	0.0592
4	0.1110	0.3333	0.0842
5	0.1404	0.1240	0.0399
6	0.1023	0.1143	0.0712
7	0.1232	0.1316	0.0841
8	0.2219	0.2362	0.1164
	Control		
1	0.0930	0.1437	0.0461
2	0.0555	0.0867	0.0368
3	0.0381	0.0595	0.0315
4	0.0381	0.0260	0.0404
5	0.0366	0.0366	0.0325
6	0.0381	0.0484	0.0526
7	0.0366	0.0430	0.0342
8	0.0459	0.0467	0.0330

Appendix B-3

Standard errors of the log transformed data of latencies for mount, intromission, and ejaculation for only those males exhibiting an ejaculatory response in Experiment 1, Figure 5.

Trial	Mount	Intromission	Ejaculate
	Odor-Sex/Ill Group		
1	0.1871	0.1499	0.0490
2	0.1036	0.1691	0.0809
3	0.1499	0.1499	0.0719
4	0.0996	0.0996	0.0700
5	0.2720	0.2720	0.1267
6	0.3042	0.3042	0.1888
7	0.1542	0.1624	0.0461
8	0.1136	0.1149	0.1219
	Sex/Ill Group		
1	0.1027	0.1633	0.0649
2	0.0807	0.0663	0.0592
3	0.0607	0.0522	0.0592
4	0.1110	0.3333	0.0842
5	0.1404	0.1240	0.0399
6	0.1023	0.1143	0.0712
7	0.1350	0.1451	0.0861
8	0.1358	0.1429	0.0803
	Control		
1	0.0930	0.1437	0.0461
2	0.0555	0.0867	0.0368
3	0.0381	0.0595	0.0315
4	0.0381	0.0260	0.0404
5	0.0366	0.0366	0.0324
6	0.0381	0.0484	0.0526
7	0.0366	0.0430	0.0342
8	0.0459	0.0467	0.0330

Appendix C-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 2--odor-sex/ill group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	40	60	330	190	50	180	*
4	50	55	60	370	190	330	200	*
	965	550	475	1195	1110	1165	*	*
	25	55	*	*	*	*	*	*
10	60	65	*	*	*	*	*	*
	515	800	*	*	*	*	*	*
	20	*	*	*	*	*	*	*
12	35	*	*	*	*	*	*	*
	600	*	*	*	*	*	*	*
	10	20	35	280	*	*	*	*
13	20	25	160	*	*	*	*	*
	545	595	610	*	*	*	*	*
	15	15	55	*	*	540	*	*
26	30	15	65	*	*	540	*	*
	690	690	510	*	*	1125	*	*
	20	25	65	300	*	65	*	*
29	45	35	65	300	*	65	*	*
	480	655	420	590	*	360	*	*
	60	45	*	*	*	485	*	*
30	60	*	*	*	*	545	*	*
	605	*	*	*	*	*	*	*
	30	*	*	*	*	*	*	*
33	30	*	*	*	*	*	*	*
	595	*	*	*	*	*	*	*
	15	35	185	360	150	215	300	*
34	25	35	300	360	150	220	310	*
	550	845	1110	740	335	800	1080	*

Appendix C-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 2--sex/ill group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	5	15	5	15	15	30	20
2	15	5	15	5	15	15	30	40
	1200	470	450	540	420	300	665	650
	10	5	5	10	15	15	20	15
3	10	15	10	15	20	15	35	15
	590	485	270	185	335	215	435	485
	15	5	5	10	5	45	55	20
9	25	5	5	10	5	45	60	20
	720	290	190	200	160	220	550	420
	10	5	10	160	*	90	*	*
15	35	15	15	*	*	465	*	*
	690	605	400	*	*	*	*	*
	30	10	10	45	70	95	90	*
16	60	10	15	65	135	95	120	*
	605	435	230	355	610	230	*	*
	60	5	5	15	115	70	175	*
18	60	10	10	15	175	235	*	*
	575	465	300	210	555	525	*	*
	15	10	15	75	150	30	60	60
19	15	20	20	110	230	30	60	70
	960	575	375	430	570	170	390	670
	20	5	5	5	15	25	15	50
21	60	5	5	10	15	25	20	50
	825	415	185	110	140	265	190	570
	15	5	10	40	15	10	15	10
31	15	5	10	40	15	10	15	15
	480	375	360	665	1035	375	380	460
	45	5	30	85	260	165	90	15
32	75	15	30	85	265	165	110	20
	760	310	140	365	610	555	630	505

Appendix C-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 2--illness control.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	5	5	10	5	10	5	5
6	20	5	5	10	5	10	5	5
	410	235	170	480	170	430	210	190
	30	5	5	5	5	60	10	5
7	65	5	5	10	15	75	10	10
	1200	600	485	405	765	*	255	260
	20	10	5	5	10	15	5	5
14	25	10	5	5	10	15	5	5
	480	475	295	270	355	450	320	395
	35	5	5	5	5	5	5	10
22	60	15	5	5	30	5	10	15
	560	480	285	465	390	450	280	340
	15	5	5	15	5	25	5	5
25	15	10	10	15	5	25	5	10
	790	300	610	635	380	405	360	415

Appendix C-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 2--sex/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	30	5	10	5	5	35	5	5
8	55	5	15	5	5	35	5	10
	475	285	650	435	230	275	330	305
	20	5	5	10	10	55	5	10
17	70	5	5	10	10	195	5	15
	570	570	290	540	460	600	220	480
	10	10	5	5	5	20	10	5
20	15	10	5	5	5	20	15	5
	610	480	450	345	330	655	245	230
	15	5	5	10	5	15	5	15
23	15	15	10	15	15	15	10	15
	455	290	295	340	835	285	315	450
	35	5	5	5	10	40	5	5
24	35	10	5	10	15	40	5	5
	690	500	410	270	320	375	365	310

Appendix C-1

Raw data latency times to the nearest 5 s for mount (top), intromit (middle), and ejaculation (bottom) responses for eight trials in Experiment 2--odor/control group.

	Trial							
Rat	1	2	3	4	5	6	7	8
	10	5	5	5	5	40	5	5
1	10	15	5	5	5	70	10	5
	405	305	360	415	430	555	320	465
	25	5	5	10	5	125	10	10
5	30	5	10	15	5	135	10	15
	550	430	295	330	315	490	365	270
	10	10	5	5	10	30	5	15
27	15	10	5	10	10	200	5	15
	450	545	490	570	585	935	295	475
	35	5	10	5	5	20	5	5
28	65	5	10	5	15	50	5	5
	605	490	420	345	300	390	305	330
	15	5	5	5	15	15	10	5
35	20	10	5	5	15	15	15	10
	900	365	365	430	330	300	230	360

Appendix C-2

Standard errors of the log transformed data of latencies for mount, intromission, and ejaculation respones in Experiment 2, Figure 8.

Trial	Mount	Intromission	Ejaculate
	Odor-Sex/Ill Group		
1	0.0769	0.0533	0.0279
2	0.2274	0.2492	0.0426
3	0.2154	0.1890	0.0634
4	0.0964	0.0860	0.0367
5	0.1183	0.1183	0.0577
6	0.1737	0.1371	0.0549
7	0.1003	0.0958	0.0048
8	0.0000	0.0000	0.0000
	Sex/Ill Group		
1	0.0833	0.0969	0.0351
2	0.0381	0.0729	0.0309
3	0.0799	0.0737	0.0502
4	0.1615	0.2099	0.0919
5	0.2212	0.2295	0.0917
6	0.1219	0.1706	0.0770
7	0.2099	0.2287	0.0786
8	0.2606	0.2461	0.0547
	Control		
1	0.0536	0.0721	0.0331
2	0.0311	0.0484	0.0332
3	0.0264	0.0422	0.0374
4	0.0652	0.0499	0.0276
5	0.0422	0.0637	0.0450
6	0.0863	0.1192	0.0449
7	0.0344	0.0474	0.0200
8	0.0470	0.0534	0.0305

Appendix C-3

Standard errors of the log transformed data of latencies for mount, intromission, and ejaculation for only those males exhibiting an ejaculatory response in Experiment 2, Figure 9.

Trial	Mount	Intromission	Ejaculate
	Odor-Sex/Ill Group		
1	0.0769	0.0533	0.0279
2	0.0774	0.0861	0.0269
3	0.1066	0.1244	0.0664
4	0.0187	0.0233	0.0738
5	0.0363	0.0363	0.1840
6	0.2076	0.1701	0.1026
7	0.0000	0.0000	0.0000
8	0.0000	0.0000	0.0000
	Sex/Ill Group		
1	0.0833	0.0969	0.0351
2	0.0381	0.0729	0.0311
3	0.0799	0.0737	0.0503
4	0.1515	0.1496	0.0794
5	0.1850	0.2044	0.0897
6	0.1285	0.1530	0.0561
7	0.1097	0.1054	0.0647
8	0.1003	0.0943	0.0268
	Control		
1	0.0536	0.0721	0.0331
2	0.0311	0.0484	0.0332
3	0.0264	0.0422	0.0374
4	0.0652	0.0499	0.0276
5	0.0422	0.0637	0.0450
6	0.0882	0.1254	0.0379
7	0.0344	0.0474	0.0200
8	0.0470	0.0534	0.0305

THE ROLE OF ODOR IN LEARNED AVERSIONS TO COPULATORY BEHAVIOR IN MALE RATS

by

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B. S., Kearney State College, 1981

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Psychology

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1984

Abstract

Male rats were tested in three experiments for acquisition of an aversion to copulatory behavior by using lithium chloride induced illness. One group (odor-sex/ill) was trained with an odorous female that had been sprayed with a novel almond odor; a second group (sex/ill) was trained with a non-odorous female. In the Pilot Study and Experiment 1 illness was contingent upon an ejaculatory response, but in Experiment 2 illness followed all encounters with an estrous female. Compared with control groups, a significant copulatory aversion was exhibited by rats in the odor-sex/ill group by showing decreased ejaculatory responses, longer latencies to display all mating responses, and requiring more trials to extinction. Male rats in the sex/ill group displayed weak aversions, evident in only 30% of the males after eight trials. Results demonstrated a novel odor cue significantly potentiated a copulatory aversion in male rats.