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SNIFFING AND MOTIVATION IN THE RAT

A Dissertation Presented

By

Samuel Henry Clarke

Submitted to the Graduate School of the
University of Massachusetts in
partial fulfillment of the requirements for the degree of

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SNIFFING AND MOTIVATION IN THE RAT

A Dissertation

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October, 1970
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ABSTRACT

The relation between inspiration rate and instrumental behavior in rats was examined using fixed-interval and fixed-ratio schedules of either sucrose or brain stimulation reinforcement. In the first experiment, the inspiration rates of sucrose reinforced rats increased just before the first press of a fixed-interval trial, thus providing generality for a previous finding with brain stimulation reinforcement. In the second experiment, rats pressing on a fixed-ratio schedule, also for sucrose, demonstrated very orderly "goal gradients" in their inspiration rates while pressing. This result was replicated in a third experiment in which rats pressed on a fixed-ratio schedule for brain stimulation reinforcement. In both fixed-ratio experiments, increasing inspiration rates were associated with constant or decreasing pressing rates. The relevance of these results to classical or contiguity conditioning interpretations of instrumental or motivated behavior was discussed.

ACKNOWLEDGEMENT

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INTRODUCTION

A proposal has recently been made that principles of motivation can be translated, without loss of explanatory power, into principles of reinforcement (Bolles, 1967). Theoretical concepts which can be used to explain motivated behavior as derived from reinforcement principles have variously been termed secondary or conditional reinforcement (Mowrer, 1960), preconsummatory excitement (Sheffield, 1966), anticipatory invigoration (Cofer and Appley, 1964), incentive motivation (Bindra, 1968), etc. Reduced to essential experimental operations, all of these conceptions involve contiguous pairing of motivationally neutral stimulation with reinforcing stimulation.

As a method of investigating the effects of incentive motivational stimuli on the general activity of rats, Bindra and Campbell (1967) paired an auditory stimulus with short trains of rewarding lateral hypothalamic brain stimulation. As a result of this pairing, the auditory stimulus elicited "walking or rearing while sniffing" above control levels. The ability of the rewarding brain stimulation to generate this "positive incentive motivational" effect in contiguously paired neutral stimuli was suggested as a property common to all positive reinforcers. The generality to support this assertion came from a similar study, reported in the same year (Bindra and Palfai, 1967), in which presentation of water was paired with a neutral stimulus in thirsty rats. The "walking or rearing while sniffing" response was observationally measured in both of these experiments using a yes-or-no classification of occurrence.

A measuring technique which allows for rate assessments of the sniffing component of this incentive motivational response pattern has recently been developed by Clarke, Panksepp, and Trowill (1970). A cannula is implanted in one nasal passage of the rat. During recording sessions, a thermocouple probe is inserted into this cannula. The temperature changes associated with the respiration or sniffing cycle induce minute voltage changes in the probe. These voltage changes can be recorded on any physiograph of sufficient sensitivity and frequency response.

The first experiment using this recording technique investigated the respiration/sniffing (inspiration) rate of rats during temporal conditioning (Clarke and Trowill, in press). In this experiment, four rats received one-second trains of rewarding lateral hypothalamic stimulation every twenty seconds for one-hundred presentations a day on two successive days. During early inter-reinforcement intervals, the inspiration rate of all four animals was highest following the brain shock. With continuing temporal conditioning, inspiration rates decreased immediately following brain stimulation and became most pronounced in the temporal interval preceding stimulation. The data from this experiment are plotted for individual rats in Figure 1. Sniffing, in the range of six to nine inspirations-per-second, was a completely consistent component of the unconditional response to the rewarding brain stimulation, a finding in line with Christopher and Butter's (1968) report that sniffing and forward locomotor activity are associated with positive stimulation of hypothalamic sites.

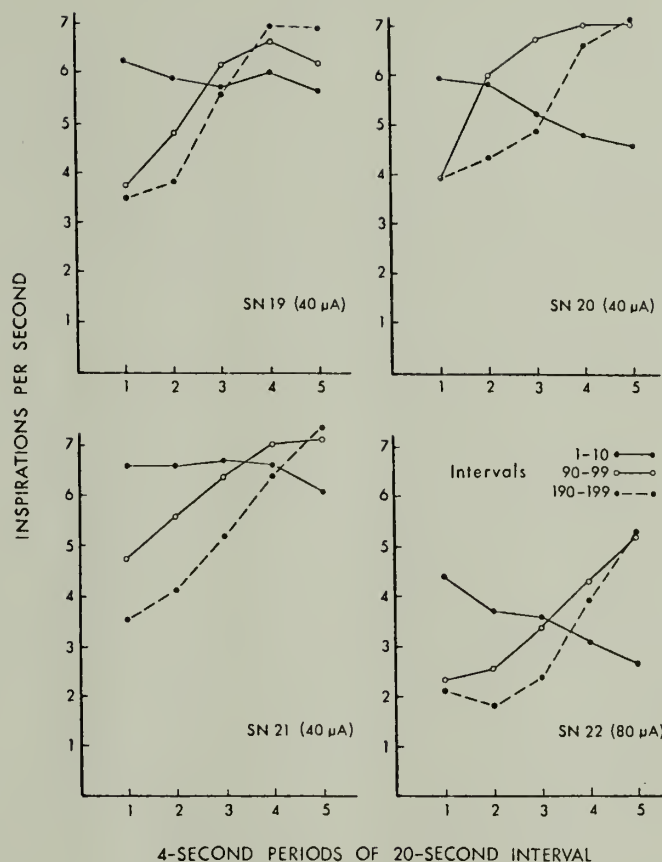


Figure 1. Individually plotted inspiration rate data of four rats during early, middle, and late stages of temporal conditioning. Each curve is the average of ten inter-reinforcement intervals. Positive brain stimulation was delivered just prior to the first four-second period, and just after the fifth four-second period of the twenty-second interval (from Clarke and Trowill, in press).

If an explanation of instrumental or motivated behavior is to be generated from classical or contiguity principles of reinforcement, it would be most useful to discover a response system in which "instrumental" patterns of response can be demonstrated when reinforcement is appropriately controlled by the experimenter rather than by the subject (Sheffield, 1965; Williams, 1965). The results seen in Figure 1 are of interest in this respect because fixed-interval "scalloping" patterns appeared in the inspiration-rate pattern when reinforcement was delivered by the experimenter in a manner analogous to its delivery during fixed-interval schedules of reinforcement.

Clarke and Trowill (in press) performed a second experiment in which the same temporal conditioning rats were run on the analogous fixed-interval 20-second (FI-20") schedule of reinforcement. Brain stimulation of the same intensity and duration was delivered through the same electrode to reinforce this lever-pressing behavior. After the rats had been trained, inspiration rate and lever pressing were recorded during a session of one-hundred fixed-interval reinforcements. Averages of ten trials (inter-reinforcement intervals) in which there was a non-reinforced lever press preceded by at least nine seconds of non-pressing, and followed by at least three seconds before reinforcement was delivered, are plotted individually for three of the rats in Figure 2. (The fourth rat did not produce a scoreable respiration/sniffing record.)

Inspection of Figure 2 shows that inspiration rate increased just prior to the first lever press of an interval, and continued to

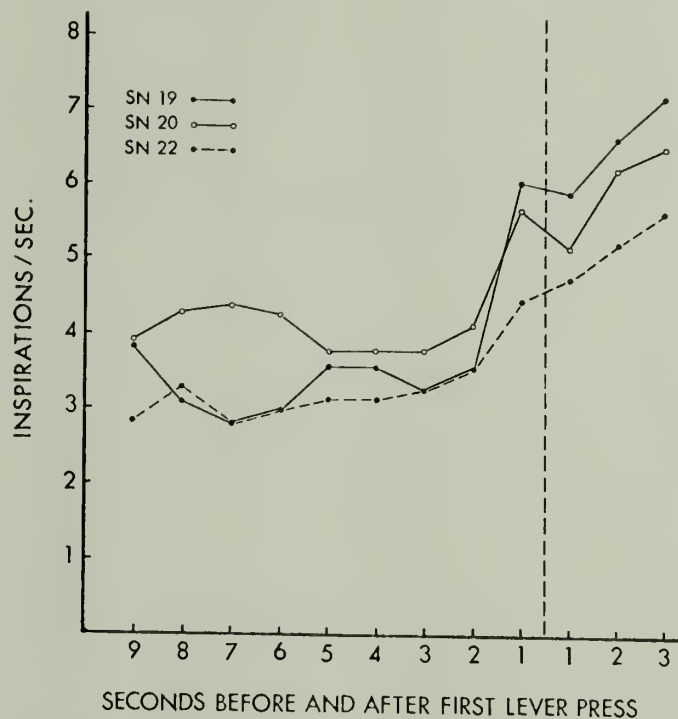


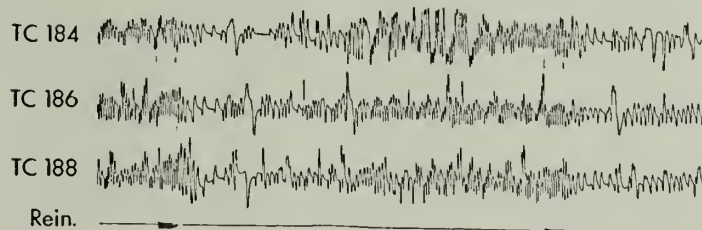
Figure 2. Individually plotted inspiration rate data of three rats responding on a fixed-interval 20-second schedule of positive brain stimulation reinforcement. Each curve is the average of ten intervals in which there was at least nine seconds of non-pressing, followed by at least three seconds of lever pressing before reinforcement was delivered (from Clarke and Trowill, in press).

increase until reinforcement was delivered. Representative records of one rat's temporal-conditioning and fixed-interval responses are presented in Figure 3.

The increase in inspiration rate immediately preceding onset of lever pressing during responding on the fixed-interval schedule suggested to Clarke and Trowill (in press) that measurement of this classically conditionable response system might have considerable relevance to theories which attempt to account for instrumental behavior in terms of classical conditioning principles of reinforcement (Mowrer, 1960; Sheffield, 1966; Stein, 1964). Stein (1964) has presented a model for self-stimulation behavior (behavior which has brain stimulation as its goal) in which excitation of the lateral hypothalamic medial forebrain bundle system (MFB) serves as both the conditionally and the unconditionally reinforcing event.

A formal model consistent with Stein's (1964) interpretation is presented in Figure 4. Motivationally neutral stimuli, when contiguously paired with reinforcement become conditionally reinforcing stimuli (A). These stimuli then have the conditional property of being able to stimulate the reinforcement system (B). Stimulation of this system causes approach and sniffing behavior (C) directed toward the conditionally reinforcing stimuli (D). Because the reinforcing effect of these conditional stimuli is a function of temporal contiguity to primary (unconditional) reinforcement, the animal is preferentially "led" into contact with the primary reinforcing stimulation by a "goal gradient" of conditional reinforcement (E). It

A. TEMPORAL CONDITIONING



B. FIXED-INTERVAL

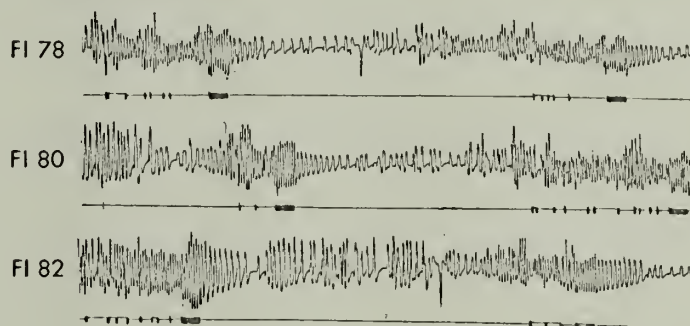


Figure 3. A. Representative physiograph records of Rat Sn19 for temporal conditioning (TC) trials 184, 186 and 188. The one-second reinforcement marks correspond to one-second trains of rewarding hypothalamic stimulation. Inspiration up. B. Representative records of the same animal responding for brain stimulation on fixed-interval (FI) trials 78, 80 and 82. The short marks on the event lines signify non-reinforced lever presses. The longer, one-second marks signify presses that were reinforced by one-second trains of rewarding brain stimulation (from Clarke and Trowill, in press).

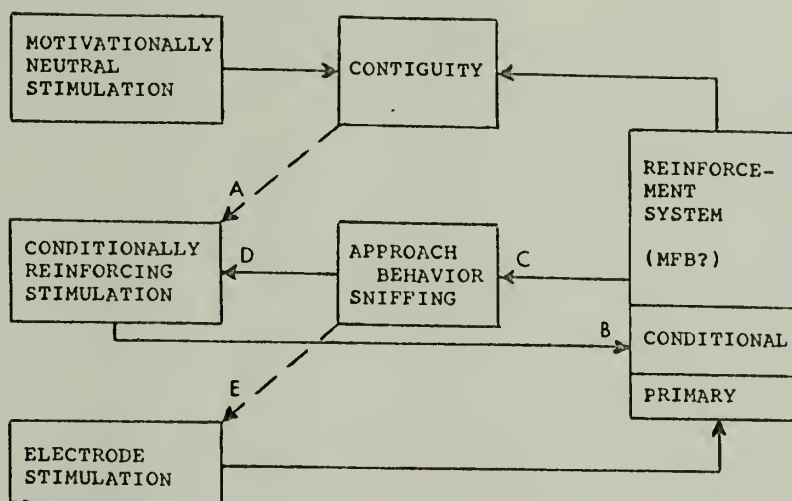


Figure 4. Formal diagram of a classical or contiguity conditioning model that accounts for instrumental behavior reinforced by positive brain stimulation. Motivationally neutral stimulation when paired with reinforcement becomes conditionally reinforcing stimulation (A). Conditionally reinforcing stimulation activates the reinforcement system (B) causing approach behavior (C) in respect to conditionally reinforcing stimulation (D). Because conditional reinforcement is a function of temporal contiguity to primary or unconditional reinforcement, the animal is "led" into contact with the unconditional or goal stimulation (E).

should be noted that activation of the reinforcement system constitutes the important response in the S - R formulation, and that inspiration-rate (sniffing frequency) reflects the degree of activation of this system in rats.

The experiments to be described in the next sections of this paper were designed (1) to investigate the generality of the fixed-interval brain stimulation finding (that inspiration frequency increased just prior to onset of lever-pressing) using sucrose as reinforcement, and (2) to discover whether inspiration frequency corresponds to the postulated gradient of conditional reinforcement while rats are emitting sequences of instrumental behavior. Both sucrose and brain stimulation were used to reinforce fixed-ratio lever pressing in the experiments designed to investigate the goal gradient hypothesis.

EXPERIMENT I

The purpose of this experiment was to investigate the generality of the findings reported by Clarke and Trowill (in press) concerning the relationship between sniffing and lever pressing for brain stimulation in the rat. In the present experiment, a 16% sucrose solution (weight/total weight) was used to reinforce fixed-interval twenty-second (FI-20") lever pressing behavior.

Methods and Materials

Subjects. Of the six approximately 120-day-old male albino rats which began this experiment, one died without recovering from surgery, one became ill, and a third did not produce a scoreable respiration-sniffing record. The experiment was completed, and data are reported, on the remaining three rats.

The animals were maintained on 14 grams of Purina Lab Chow per day until the point in the experiment at which they were pressing for more than 90 reinforcements (five-second opportunities to lick 16% sucrose) on a FI-20" schedule during a thirty-minute training session. At this time, the daily ration was increased by 2 grams per day until the number of reinforcements dropped below ninety. Without going below 14 grams per day, an attempt was made to keep the number of reinforcements between 80 and 90 for a daily thirty-minute session. By this criterion, the daily ration fluctuated between 14 and 24 grams per day for individual animals

At the beginning of the experiment, the three rats on whom data ^(is) reported weighed 538 (rat Sn25), 612 (Sn26) and 500 (Sn27) grams. At

the time of testing, their respective weights were 469, 537 and 431 grams. Water was available ad libitum in the home cage throughout the experiment.

Surgery. The procedure used for cannula implantation was a slight modification of that reported by Clarke, Panksepp, and Trowill (1970). The rats were anesthetized with Diabutal (40mg/kg) and placed in a stereotaxic headholder. The skull was bared from the tip of the snout back to the parietal bones. Five jewelers screws for anchoring the cap were fixed to the skull, one in a nasal bone, and one in each of the frontal and parietal bones. A 5/64th inch hole was then drilled in the contralateral nasal bone 1 to 2mm back from its anterior end. After removing the periosteum in the vicinity of the hole with a cautery, a 1/2in #15 gauge cannula was positioned such that its lower extent was even with the lower plane of the nasal bone. Dental cement was then molded around the screws and the cannula to form a cap extending along the anterior two-thirds of the animal's skull. A small hook-eye was placed in the cap to provide a means of securing the thermocouple probe within the cannula during recording sessions.

Apparatus. Training and testing were done in a 22x33x40cm sound attenuated chamber. Masking noise (70dB) was provided by a BRS-Foringer AG901 sound generator.

The 16% sucrose solution was delivered through a 1.5cm diameter opening on the left side of the chamber, 5cm from the rear corner and 6cm above the wire mesh floor. The mechanism of delivery consisted of a 100ml drinking bottle and stainless steel drinking tube mounted in

such a way that activation of a solenoid would pull the drinking tube into the chamber. The masking noise was terminated whenever the drinking tube was in the chamber thus providing, along with the solenoid sound, an added stimulus change to signal opportunity for reinforcement.

A rat lever (Lehigh Valley 1352) was mounted on the rear wall of the chamber, 5cm from the left corner and 3cm above the floor. The lever protruded only 1cm into the chamber to prevent the thermocouple probe assembly from becoming tangled.

The respiration/sniffing response was recorded on a physiograph (E&M Instrument Company) as described in the communication of Clarke, Panksepp, and Trowill (1970). Licking was also recorded on the physiograph using an E&M Impedance Pneumograph connected to the relay contacts of a Grason-Stadler Drinkometer. In this way, a downward deflection of the recording pen resulted whenever the animals made contact with the drinking tube. Lever pressing and delivery of reinforcement were recorded on the Event Marking channel of the physiograph.

Procedure. After three days at 14 grams of Purina Lab Chow per day, the rats were given two daily one-hour sessions of habituation to the chamber during which the sucrose tube was continually available. The rats were then shaped to approach the tube whenever the sound of the solenoid and the offset of the masking noise signalled the presense of the sucrose tube in the chamber. Once this stimulus change had become reliable discriminatory stimulation for tube approach, it was used as conditional reinforcement to shape lever-pressing behavior. Once the

rats had learned to lever press, they were given a 30-minute session of continuous reinforcement (a five-second opportunity to lick sucrose consequent to each lever press). From this point on, daily sessions were 30 minutes.

Over the next five days, the rats were gradually trained to press on FI-20" schedules. A FI-5" schedule was used on the first of these days, and was followed by intervals of non-reward set respectively for each subsequent day at 7.5, 10, 15 and 20 seconds.

Surgery for cannula implantation was then performed. Following surgery, the rats were given 20 further days of training on the FI-20" schedule. This extensive amount of further training was needed because it was observed that the animals gnawed on the lever while they were pressing. Previous experience with other animals had shown that gnawing eliminated sniffing in rats.

Sniffing, licking, and lever-pressing were recorded for 30 minutes on the twenty-first post-surgery session. The number of reinforcements accumulated during this session by rats Sn25, Sn26 and Sn27 were 83, 92 and 81 respectively.

Results

The results of this experiment were subjected to the same analysis as were the results of the fixed-interval brain stimulation experiment (Clarke and Trowill, in press). The last ten intervals for each rat in which there was a non-reinforced lever press, preceded by at least nine seconds of non-pressing, and followed by at least three seconds of non-reinforcement, were hand-scored for inspiration rate on a second-by-

second basis. The averaged data for each rat is presented in Figure 5. As in the fixed-interval brain stimulation experiment, inspiration rate increased just before onset of pressing. For rat Sn25, nine out of the ten scores for the second preceding onset of pressing were higher than the average of the total (90) pre-pressing scores for this animal. For the other two rats, ten out of ten scores for this last second before pressing were higher than the respective averages of their total pre-pressing scores. The unaveraged data from this experiment are given in Appendix A.

Unlike the results of the brain stimulation experiment, sniffing did not continue to increase so markedly during the pressing portion of the interval. This comparison is illustrated in Figure 6, which represents data averaged across the three rats in each experiment. Despite the extensive post-surgery training given in the present experiment, the sucrose rats continued to gnaw at the lever. Since sniffing appears to be incompatible with gnawing in the rat, such lack of parallelism as seen in Figure 6 may have resulted from the greater amount of gnawing (informal observation) done by the sucrose animals.

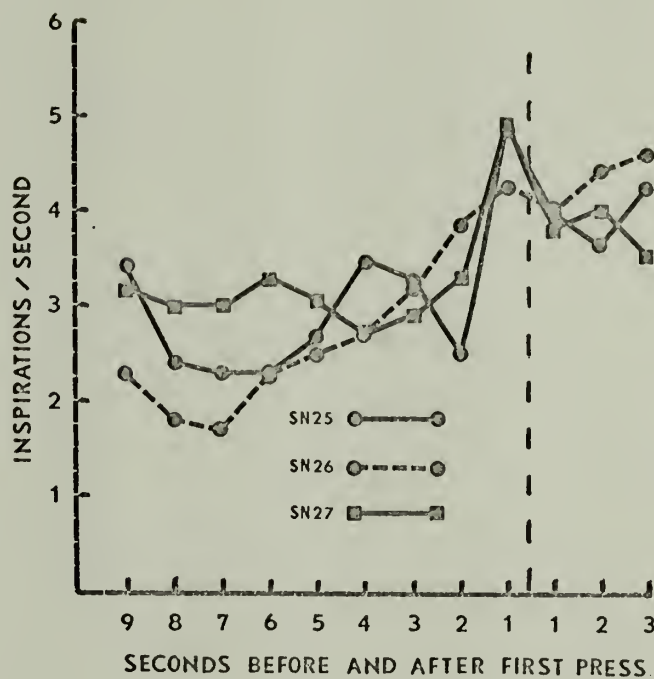


Figure 5. Individually plotted inspiration rate data of three rats responding on a fixed-interval twenty-second schedule of sucrose reinforcement. Each curve is the average of ten intervals in which there was at least nine seconds of non-pressing, followed by at least three seconds of lever pressing before reinforcement was delivered.

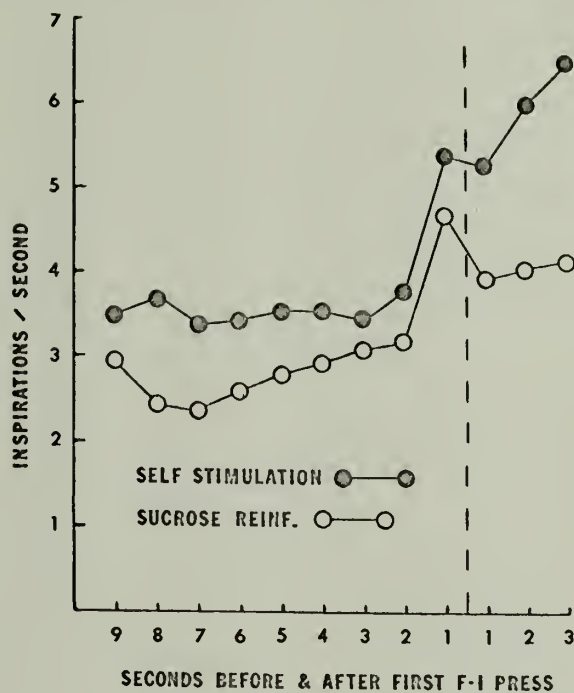


Figure 6. Mean inspiration rate data of three rats responding for brain stimulation (Clarke and Trowill, in press), and three rats responding for sucrose, on a fixed-interval 20-second schedule of reinforcement.

EXPERIMENT II

If the contiguity model presented in Figure 4 is essentially sound, and if sniffing frequency adequately reflects the amplitude changes in conditional reinforcement postulated by this model, then rats pressing on fixed-ratio schedules of reinforcement should demonstrate positive gradients of inspiration rate while emitting fixed-ratio series of lever presses. In this experiment, a 16% sucrose solution (weight/total weight) was used to reinforce fixed-ratio ten (FR-10) lever pressing behavior.

Methods and Materials

Subjects and Surgery. Of the six approximately 100-day-old male albino rats that began this experiment, one failed to learn to press the lever and two did not produce scoreable respiration/sniffing records when tested. The experiment was completed, and data are reported, on the remaining three animals. As in Experiment I, the rats were maintained on between 14 and 24 grams of Purina Lab Chow per day, adjusted to keep the number of reinforcements between 80 and 90 per daily 30-minute training session. At the beginning of the experiment, the three rats weighed 475 (rat Sn29), 490 (Sn33), and 524 (Sn34) grams. At the time of testing, when they were approximately 150-days-old, their respective weights were 476, 478 and 487 grams. Water was available ad libitum in the home cage throughout the experiment.

Surgery for cannula implantation was performed at the point in the experiment when the rats were performing on the FR-10 schedule. The surgical procedure was identical to that used in Experiment I.

Apparatus. The apparatus used in this experiment was the same as that used in Experiment I except for a change in levers. In this experiment, the small Lehigh Valley Rat Lever was replaced by a larger Gerbrands (G6312) Rat Lever. Substitution of this larger lever effectively reduced the amount of lever gnawing in two of the three rats.

Procedure. The rats were trained to press the lever as in Experiment I. They were then given six days of fixed-ratio training during which time the schedule was gradually increased from a FR-2 through FRs 3, 5, 7, 9 and finally ten. Surgery for cannula implantation was then performed.

Following surgery, the rats were given 29 further days of FR-10 training. This amount of training was used to eliminate premature approaches to the site of reinforcement during a fixed-ratio series. Lever pressing, licking, and respiration/sniffing behavior were recorded for 40 minutes on the 30th post-surgery day.

Results

The physiograph records from this experiment were, ignoring the first ten fixed-ratio series (trials), scored (1) for the duration of the post-reinforcement pause (the interval between reinforcement and the onset of pressing for the following reinforcement) and (2) for the duration between onset of each series of fixed-ratio presses and the delivery of reinforcement. The longest possible post-reinforcement pause duration that allowed for selection of three blocks of ten trials, grouped according to pressing duration, was determined for each rat. No trial was analysed in which there was an interval of three or more seconds between any two presses of a series.

For rats Sn33 and Sn34, the post-reinforcement pause segment which allowed analysis by the above criterion was two seconds. For rat Sn29, this duration was one second. This last rat rarely pressed the lever; rather it shook the lever with mouth and front paws and produced "pressing" rates in the neighborhood of 6 microswitch contacts per second.

The data from the two rats that pressed the lever are plotted separately for each rat on a second-by-second time scale in Figure 7. For comparison, inspiration-per-second ("sniffing") data are plotted above presses-per-second ("pressing") data. The right-most extension of each curve gives the particular duration of pressing illustrated by that curve. Both rats show very orderly and altogether remarkable "goal gradients" in the respiration/sniffing response while they are engaged in fixed-ratio series of lever presses. It can also be noted that, while their sniffing rates increased, their pressing rates remained relatively constant (Sn34) or decreased (Sn33). The data from the individual trials associated with each curve are listed in order, beginning with those trials closest to the end of the recording session, in Appendix B.

In contrast to the behavior of both the brain stimulation and sucrose fixed-interval rats, these fixed-ratio animals often began pressing the lever with little or no "forewarning" given by their sniffing responses. This observation, difficult to make-out from Figure 7 because the analysable post-reinforcement pause duration was so brief, was more apparent in the physiograph records. Representative records

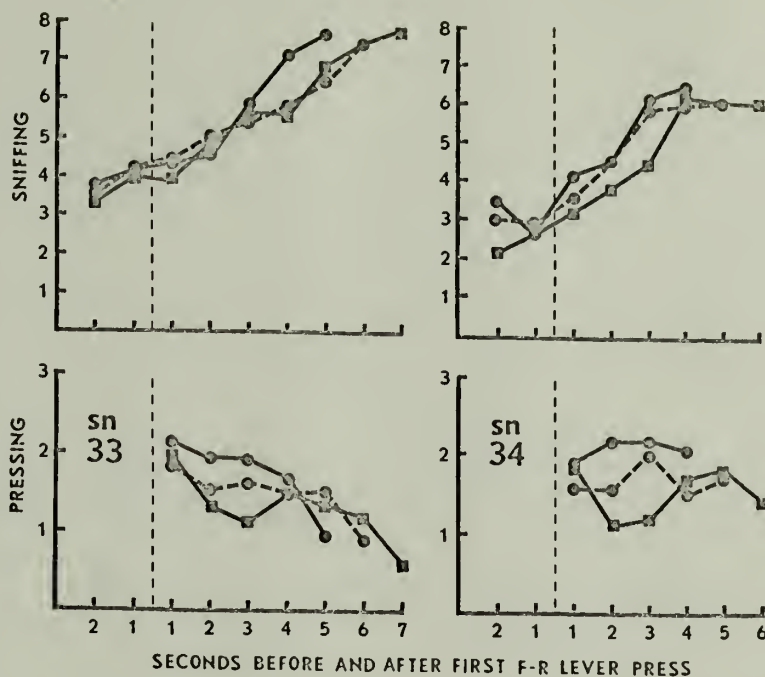


Figure 7. Individually plotted inspirations-per-second (sniffing) and presses-per-second (pressing) data of two rats responding on a fixed-ratio ten schedule of sucrose reinforcement. Each curve is the average of ten fixed-ratio trials. The right-most extension of each curve gives the pressing duration associated with that curve.

of rat Sn33 are given in Figure 8. The data from the lever-shaking Sn29 is presented on a 1/2second-by-1/2second time scale in Figure 9, along with the data of another lever-shaker from the experiment next to be considered.

FIXED-RATIO TEN

SUCROSE

Sn33

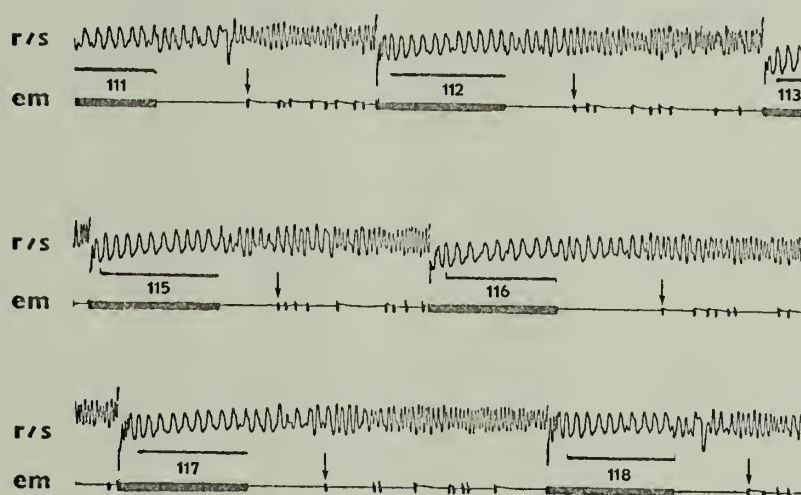


Figure 8. Representative respiration/sniffing (r/s) records of rat Sn33 responding for sucrose on a fixed-ratio ten schedule of reinforcement. The short marks on the event marking (em) channel signify lever presses. The longer, five-second, marks signify presses reinforced with five-second opportunities to lick sucrose. The thin lines above the numbers for each reinforcement signify the onset and duration of licking. Inspiration up.

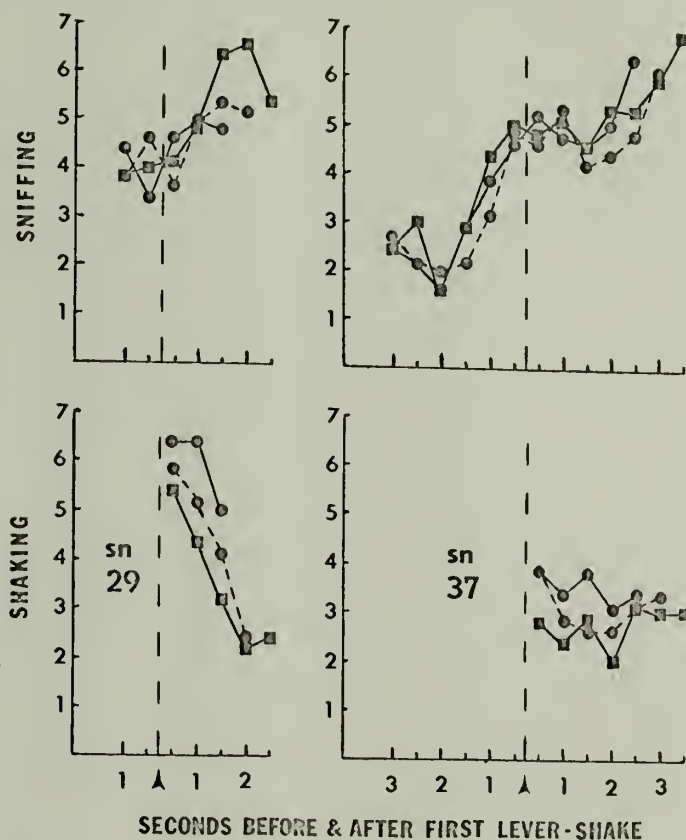


Figure 9. Individually plotted inspirations-per-second (sniffing) and microswitch contacts-per-second (shaking) data. Rat Sn29 was rewarded on a fixed-ratio schedule with sucrose; rat Sn37 was rewarded on the same schedule with brain stimulation. Each curve is the average of ten fixed-ratio trials. The right-most extension of each curve gives the "shaking" duration associated with that curve.

EXPERIMENT III

In this final experiment, three more rats were trained on a fixed-ratio ten (FR-10) schedule of brain stimulation rather than sucrose reinforcement. It has been shown thus far that the respiration/sniffing response pattern of rats pressing on fixed-interval twenty-second schedules is similar for both brain stimulation and sucrose reinforcement. This experiment asks whether the inspiration-rate goal gradients seen in the fixed-ratio sucrose rats will also appear in the sniffing response of rats pressing on a fixed-ratio schedule for lateral hypothalamic stimulation.

Within the lower limit required for consistent fixed-ratio behavior, the level of reinforcing brain stimulation was adjusted to produce maximally long post-reinforcement pause durations, thus allowing for an improved investigation of inspiration frequency during this important period of response initiation.

Methods and Materials

Subjects and Surgery. Four mature male albino rats which displayed lever pressing behavior for stimulation of monopolar electrodes aimed at the lateral hypothalamus were selected for this experiment. Stereotaxic coordinates of 1.4mm posterior to bregma, 1.6mm lateral to the midline, and 8.9mm ventral from the skull top, were used for the electrode placements in conjunction with the brain atlas of Pellégrino and Cushman (1967). Each rat was also implanted with a nasal cannula as in Experiments I and II. This second operation was a failure for one rat.

Throughout the experiment, the animals were maintained on ad libitum

Purina Lab Chow and water in the home cage. At the end of the experiment, the three rats on whom data are reported were approximately 120-days-old and weighed 400, 436 and 480 grams.

Apparatus. The chamber and lever used were the same as in the fixed-ratio sucrose experiment (Experiment II). Brain stimulation consisted of 1.0-second trains of 60 cycle sine waves of a constant current intensity, individually established for each animal, that just maintained consistent FR-10 lever pressing. Current intensity was monitored with an AC microammeter in series with the rat. For rats Sn35, Sn37 and Sn38, this current intensity was 30, 80 and 40 microamperes respectively. Lever pressing and respiration/sniffing frequency were measured as described in Experiment I.

Procedure. Two days following surgery for nasal cannula implantation, the rats began training on fixed-ratio schedules. During the first three days of training, the schedule was gradually increased (FR-3, FR-5, FR-7) to the FR-10 schedule. Each daily session was one-hour. On the fourth day, the rats were given one further hour of FR-10 training. On day five, lever pressing and respiration/sniffing behavior were recorded for 45 minutes. During this recording session, rats Sn35, Sn37 and Sn38 received 109, 196 and 153 reinforcements respectively.

Results

The results of this experiment were subjected to the same analysis as were the results of the FR-sucrose study (Experiment II). Ignoring the first ten trials, the physiograph records were scored for the duration of the post-reinforcement pauses, and for the time taken to

complete each series of FR lever presses from onset of pressing to delivery of reinforcement. The longest possible post-reinforcement pause duration that allowed for selection of three blocks of ten trials, grouped according to pressing duration, was determined for each rat. No trial was analysed in which there was an interval of three or more seconds between any two presses of a series.

For rats Sn35 and Sn38, the post-reinforcement pause segments which allowed for analysis by the above criterion were four and three seconds respectively. For rat Sn37, this duration was three seconds. This last rat was another lever-shaker; its data are presented on a 1/2second-by-1/2second time scale in Figure 8, along with the data from the similarly behaving rat in the FR-sucrose experiment.

The sniffing and pressing data (in inspirations and presses per second) from the two lever-pressing rats are plotted on a second-by-second time scale in Figure 10. As in the previous two figures, the right-most extension of each curve gives the particular pressing duration associated with that curve. Just as seen in the FR-sucrose experiment (Figure 7), both rats demonstrate very orderly "goal gradients" in the sniffing response while they are emitting fixed-ratio series of lever presses. In contrast to the FR-sucrose animals, these brain stimulation reinforced rats also show clear gradients of a positive slope in the period prior to the first press of a fixed-ratio series. The unaveraged data for the curves of the Figure 9 animals are given in Appendix C, again in reverse order of occurrence.

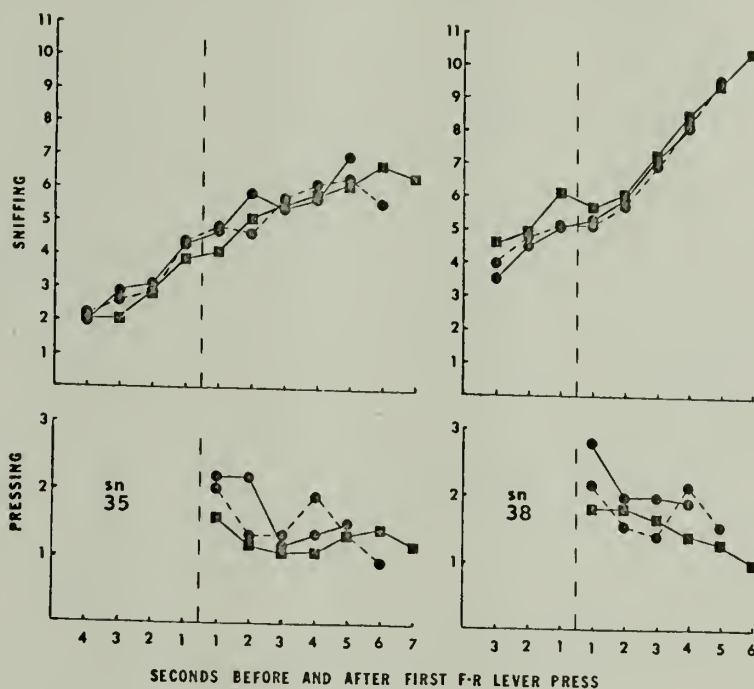


Figure 10. Individually plotted inspirations-per-second (sniffing) and presses-per-second (pressing) data of two rats responding on a fixed-ratio ten schedule of brain stimulation reinforcement. Each curve is the average of ten fixed-ratio trials. The right-most extension of each curve gives the pressing duration associated with that curve.

DISCUSSION

The pattern of inspiration frequency emitted while rats were pressing on fixed-interval schedules for sucrose reinforcement (Experiment I) is primarily to be seen as a demonstration of the generality of similar patterns emitted by rats pressing on the same schedule for positive brain stimulation (Clarke and Trowill, in press). This finding, along with the similarity of inspiration-rate patterns seen in rats pressing on fixed-ratio ten schedules for both sucrose (Experiment-II) and brain stimulation (Experiment III) reinforcement, is supportive of Bindra and Campbell's (1967) suggestion that "a feature common to reinforcers may be the ability to generate or condition incentive-motivational properties...."(p.375).

While the remarkable goal gradients seen in the inspiration rates of rats pressing on fixed-ratio ten schedules does not prove the validity of classical conditioning interpretations of instrumental behavior, this demonstration certainly establishes the respiration/sniffing response as particularly worth investigating in the light of such interpretations. These gradients were seen during both the response initiation period and after the onset of pressing in all three brain stimulation animals, but in only one of the sucrose animals. This difference can possibly be rationalized by noting the shorter post-reinforcement pauses of the latter rats, and by suggesting that during this part of the interval, sucrose remained in the mouth and was being swallowed. Swallowing may not be compatible with increases in inspiration rate.

Further consideration of the matter of response initiation shows, however, that this problem will not be easily solved. The behavior associated with the increase in inspiration rate just prior to onset of pressing during fixed-interval responding for brain stimulation (Clarke and Trowill, in press) is worth describing in this regard. Very typically, these rats faced away from the lever immediately after brain stimulation and, quite often, lay down. This position was then maintained throughout most of the inter-reinforcement interval. Toward the end of this interval, their vibrissae began suddenly to twitch while, at the same time, their snouts were raised. Almost if not immediately thereafter, the rats rose and returned to the lever. Vibrissae protraction and retraction is a sure indication that a rat is sniffing (Welker, 1964).

The point of this description is that the lever press (as measured by a microswitch closure) cannot realistically be used to define initiation of instrumental behavior; the rising and returning to the lever are equally components of this behavior. Even if instrumental behavior is under the control of conditionally reinforced or classically conditioned incentive-motivational states, the time lag between activation of the motivational system and the appearance of instrumental behavior may well be determined by the time nervous tissue takes to propagate impulses. When an assumed peripheral index of the motivational state is used, the response of this system may appear simultaneously with the initiation of instrumental behavior. It is therefore conceivable that the final defense of a contiguity or classical conditioning interpre-

tation will have to be made on other grounds, eg., in the name of parsimony.

While an extensive discussion of the role of deprivation in relation to incentive processes is not appropriate to this paper (because deprivation was not herein studied systematically), many theorists have concluded that the deprivation operation does not directly influence behavior, but rather exerts its drive-like effects through incentive mechanisms (Bindra, 1968; Bolles, 1967; Cofer and Appley, 1964). In the case of feeding, food deprivation makes food reinforcing while, at the same time, it makes stimuli previously associated with food conditionally reinforcing (or motivating). Trowill, Panksepp, and Gandelman (1969) have concluded from their review of the brain stimulation literature that positive brain stimulation is ideal for investigations of incentive-motivational processes because here the deprivation operation is not needed to generate motivated behavior. It may, therefore, be heuristically valuable to attribute similarities between deprivation-induced and brain stimulation-induced behavior to the action of incentive mechanisms.

The positively sloped gradients in the respiration/sniffing rates of rats pressing on fixed-ratio schedules has obvious relevance to the approach (strength-of-pull) gradients produced by rats in runways when rewarded by conventional reinforcers (Brown, 1948; Miller, 1959) and by positive brain stimulation (Sonderegger and Rose, 1970). Miller (1959) has stressed the need for an independent measure that would "index ... rapid changes in fear or general activation" (p.235) while rats are in

the approach/avoidance runway situation. While inspiration rate may not follow the predicted course of these intervening variables, it does change rapidly and it does appear to be relevant to the approach/avoidance runway experiment.

Clarke and Trowill (in press), following the lead of Bindra and Palfai (1967) have suggested that inspiration rate may provide a measurable response that indexes motivational states across the entire spectrum of positive to negative incentive conditions. While as yet unsystematically studied, inspiration rates persistently as low as one-half cycle per second have been seen during pilot observations when rats were anticipating or receiving strong tail shock. Bindra and Palfai (1967) reported that, whereas stimuli paired with positive reinforcement increased "walking or rearing while sniffing," cues paired with aversive reinforcement (foot shock) decreased the incidence of this behavior as compared with control levels.

It is noteworthy, in regard to this idea of a continuum, that inspiration rate does not show abrupt changes at identifiable points on its frequency spectrum (Clarke and Trowill, in press; cf. also Figures 3 and 8). An estimation of the range of sniffing frequency (5-11 Hz) has, however, been made on the basis of vibrissae, nares, and head movements (Welker, 1964).

In the concluding paragraphs of this paper, the question should be asked: why expect inspiration rate in the rat to be any better a measure of incentive than has been salivation in the dog? Results of fixed-ratio experiments have shown that lever pressing very reliably precedes

salivation (Kintsch and Witte, 1962; Williams, 1965). Ellison and Konorski (1964) have also "separated" lever pressing and salivation with a procedure in which a visual stimulus served as a discriminative cue for a fixed-ratio series of lever presses that culminated with an auditory stimulus paired with food. Salivation occurred only to the auditory stimulus.

Sheffield (1965) has understood the failure of salivation to provide a measure relevant to incentive processes by pointing to what Pavlov (1927) called "inhibition of delay." In other words, the dog makes a temporal discrimination and salivation comes to occur closer and closer to the time of food presentation. With sufficient training, conditioned salivation can only be demonstrated by omitting the reward.

Egger and Miller (1963), on the evidence of a number of studies (cf. also Egger and Miller, 1962) dealing with secondary or conditional reinforcement, conclude that "the arrival of information about primary reward may be the only occasion when reinforcement occurs" (p.132). Ignoring certain problems with this statement, the point is well-made that the effects of reinforcement appear to be at least partly perceptual in character. Thus the process of incentive (reinforcement-dependent) motivation might better be reflected in a perceptual (eg., sniffing) rather than digestive (eg., salivation) response system.

Clearly, experiments using longer fixed-ratio schedules and experiments investigating the "inhibition of delay" problem need to be done before inspiration rate can be used as a compelling index of incentive motivation. Experiments also need to be done which more directly

relate inspiration frequencies to magnitudes of conditional reinforcement. The experiments reported in this paper can be seen as the beginning of a program of research along these lines.

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Appendix A

Unaveraged data for rats Sn25, Sn26, and Sn27. The data for each animal are listed in order starting backward from the end of the recording session.

Seconds before and after first fixed-interval lever press

	9	8	7	6	5	4	3	2	1	1	2	3
<u>Sn25</u>	7	3	2	1	2	3	2	2	6	3	4	8
	5	5	3	2	4	6	3	1	4	3	5	1
	1	2	2	2	1	2	5	2	5	5	4	7
	2	2	4	3	5	5	3	3	6	7	5	7
	2	1	4	5	4	6	4	2	4	4	3	2
	6	2	1	1	2	1	5	2	3	2	2	4
	1	2	3	2	3	3	3	2	6	5	5	2
	3	2	0	2	2	2	3	2	4	2	2	1
	2	2	2	2	1	2	2	3	5	4	4	5
	5	3	2	3	3	5	3	6	6	5	3	6
<u>Sn26</u>	2	1	1	2	2	4	1	2	3	4	5	5
	3	2	2	4	2	2	4	3	5	3	5	5
	1	2	1	2	1	1	2	4	3	3	5	4
	3	3	2	2	3	4	5	5	3	4	5	4
	3	1	2	1	2	3	2	4	4	5	2	5
	2	1	0	2	1	2	3	3	5	7	5	5
	2	2	1	2	3	5	3	4	5	3	4	4
	2	1	2	2	3	3	3	5	3	2	5	4
	1	2	3	3	2	1	6	4	5	6	5	7
	4	3	3	3	6	2	3	5	7	4	4	4
<u>Sn27</u>	3	2	2	4	5	3	2	5	5	4	4	2
	6	7	6	5	1	3	4	5	5	4	5	5
	4	1	3	2	2	1	2	2	4	2	2	4
	2	3	2	2	2	3	0	2	5	2	4	3
	1	2	2	2	3	2	2	4	4	3	2	1
	6	6	5	4	5	5	3	1	4	5	4	6
	3	2	2	3	1	1	1	4	5	4	7	5
	2	4	3	4	2	1	4	3	6	5	6	3
	2	1	2	3	4	5	4	1	6	4	3	5
	3	2	3	4	5	6	5	6	6	5	4	2

Appendix B

Unaveraged data for Rats Sn33 and Sn34. The data are listed in backward order. (Corresponding presses are in parentheses.)

Sn33: Seconds before and after first fixed-ratio lever press

2	1	1	2	3	4	5	6	7
3	3	4(3)	5(0)	5(2)	7(2)	8(0)	8(1)	8(1)
3	3	5(2)	7(1)	8(1)	7(2)	7(1)	8(2)	7(0)
3	3	2(3)	4(1)	5(1)	5(1)	6(2)	7(1)	8(0)
2	4	3(1)	4(1)	5(1)	5(1)	6(2)	7(2)	7(1)
5	4	4(2)	6(1)	5(0)	6(2)	6(2)	6(1)	9(1)
5	3	4(2)	4(2)	6(1)	2(0)	5(1)	7(3)	9(0)
3	4	4(2)	5(1)	6(2)	8(1)	9(1)	8(1)	8(1)
5	5	4(2)	3(1)	4(1)	5(3)	7(2)	6(0)	6(0)
2	5	4(2)	4(2)	3(2)	3(2)	5(0)	9(0)	9(1)
3	6	5(1)	5(3)	9(0)	8(1)	9(2)	9(1)	8(1)
3	4	4(1)	4(2)	5(3)	5(0)	6(2)	9(0)	
4	2	3(3)	5(1)	3(2)	4(0)	4(2)	7(1)	
3	5	5(3)	5(0)	5(1)	7(2)	6(1)	8(1)	
3	4	3(1)	6(1)	5(2)	5(2)	7(2)	7(1)	
4	2	3(2)	4(2)	6(1)	3(1)	7(2)	7(1)	
2	4	4(2)	6(2)	5(0)	6(2)	8(1)	7(2)	
3	5	4(1)	5(2)	6(2)	8(2)	8(2)	7(0)	
5	5	6(1)	5(2)	7(2)	7(2)	9(1)	7(1)	
6	5	7(2)	5(1)	6(1)	5(2)	7(1)	8(1)	
3	6	5(2)	6(2)	6(2)	7(2)	7(1)	8(0)	
3	3	4(3)	3(1)	5(1)	4(0)	6(3)		
4	5	4(1)	4(3)	8(2)	9(2)	8(1)		
3	3	4(2)	5(2)	6(3)	9(2)	8(1)		
4	4	4(2)	6(1)	6(2)	9(3)	8(0)		
5	6	6(2)	5(1)	6(2)	6(2)	7(1)		
6	4	5(3)	4(3)	6(2)	7(1)	9(0)		
3	3	4(2)	6(3)	6(1)	8(1)	9(1)		
3	4	3(2)	3(1)	4(2)	5(2)	5(0)		
4	3	4(2)	5(2)	6(2)	8(2)	8(1)		
3	6	5(2)	4(2)	6(2)	7(2)	8(1)		

Sn34: Seconds before and after first fixed-ratio lever press

2	1	1	2	3	4	5	6
1	2	4(2)	5(2)	6(1)	7(2)	5(0)	7(2)
4	4	5(2)	6(0)	7(2)	6(1)	7(2)	7(2)
5	2	3(2)	2(1)	4(0)	6(2)	7(3)	7(1)
2	3	2(1)	1(0)	4(2)	6(2)	7(2)	6(2)
2	2	3(2)	5(3)	7(0)	8(1)	5(1)	7(2)
1	3	2(1)	4(2)	6(2)	6(1)	5(3)	7(0)
2	2	2(1)	2(0)	3(2)	5(1)	7(3)	4(2)
1	1	4(2)	3(1)	4(0)	6(3)	5(2)	4(1)
1	5	3(3)	6(1)	6(1)	6(2)	7(1)	6(1)
3	2	4(2)	4(1)	4(2)	6(2)	6(1)	6(1)

2	1	4(1)	3(1)	3(2)	6(1)	6(2)
1	3	1(1)	6(0)	7(4)	6(2)	7(2)
3	3	5(3)	5(2)	6(2)	5(0)	6(2)
6	5	7(2)	6(2)	7(1)	6(2)	6(2)
4	3	2(2)	5(3)	7(1)	7(2)	5(1)
3	3	2(1)	4(4)	5(1)	5(0)	7(2)
2	2	3(1)	3(1)	5(3)	5(2)	6(2)
3	2	4(2)	6(3)	7(2)	7(1)	5(0)
4	4	5(1)	3(0)	7(3)	7(2)	7(2)
2	2	3(2)	4(0)	5(2)	6(3)	6(2)

3	2	6(2)	7(3)	7(2)	6(1)
3	3	5(2)	5(3)	7(2)	6(2)
3	2	4(2)	7(3)	6(2)	6(1)
3	3	3(2)	3(2)	4(2)	7(2)
4	2	3(2)	2(1)	6(3)	5(2)
3	2	2(1)	4(1)	6(2)	7(4)
4	2	5(3)	4(2)	7(2)	7(2)
4	3	7(1)	5(3)	7(2)	7(3)
6	4	3(2)	4(2)	5(2)	6(3)
2	3	3(2)	4(2)	6(3)	8(1)

Appendix C

Unaveraged data for Rats Sn35 and Sn38. The data are listed in backward order. (Corresponding presses are in parentheses.)

Sn35: Seconds before and after first fixed-ratio lever press

4	3	2	1	1	2	3	4	5	6	7
3	2	1	5	4(1)	6(2)	6(1)	6(2)	5(0)	8(2)	8(1)
1	3	3	5	4(1)	5(2)	5(0)	6(2)	8(1)	7(2)	4(1)
2	2	1	4	3(2)	5(0)	7(1)	5(1)	5(2)	7(1)	8(2)
1	2	2	3	4(2)	5(0)	5(2)	4(1)	6(1)	6(2)	4(0)
2	2	4	4	4(1)	6(2)	6(1)	5(1)	8(2)	8(0)	6(1)
4	2	5	5	5(3)	4(1)	6(1)	7(1)	4(1)	5(1)	8(1)
2	2	2	3	4(1)	5(2)	6(2)	4(0)	8(1)	8(2)	7(2)
2	2	4	5	4(2)	6(2)	5(1)	7(1)	6(1)	7(1)	6(1)
2	2	3	3	6(2)	5(0)	5(1)	8(1)	6(2)	5(1)	7(2)
2	2	3	2	3(1)	4(1)	4(1)	6(1)	6(2)	6(2)	6(1)
2	1	2	2	3(1)	5(1)	6(3)	5(2)	7(1)	8(1)	
2	2	3	5	4(2)	5(1)	4(1)	7(3)	7(1)	7(1)	
1	3	2	4	4(2)	5(1)	5(1)	4(1)	7(1)	8(3)	
2	3	3	5	6(2)	3(1)	6(1)	8(2)	7(2)	4(0)	
3	3	4	4	5(3)	4(2)	4(0)	6(1)	5(2)	3(0)	
3	2	2	3	5(1)	5(1)	5(1)	6(2)	4(2)	4(1)	
3	3	4	6	5(3)	5(1)	7(1)	7(2)	7(1)	6(1)	
2	2	3	5	5(1)	4(2)	6(2)	7(2)	7(1)	6(1)	
2	4	3	5	6(2)	5(2)	7(2)	7(2)	6(1)	5(0)	
2	3	3	5	6(3)	5(1)	7(1)	5(2)	7(1)	5(1)	
2	2	2	5	5(3)	4(2)	3(0)	5(1)	7(2)		
2	2	5	3	5(2)	8(4)	4(1)	5(1)	9(1)		
2	2	2	5	3(2)	5(2)	7(1)	6(2)	6(2)		
2	3	3	4	5(3)	5(2)	5(1)	6(1)	4(1)		
2	4	4	5	6(2)	7(1)	6(1)	5(2)	8(2)		
2	4	3	2	4(2)	6(2)	4(2)	5(1)	8(2)		
2	1	3	5	6(2)	5(3)	5(1)	7(2)	7(1)		
1	5	6	4	5(2)	6(2)	6(1)	7(1)	8(2)		
2	2	1	5	3(3)	6(1)	7(3)	5(1)	5(1)		
3	4	2	5	5(1)	7(3)	7(1)	6(1)	8(1)		

Sn38: Seconds before and after first fixed-ratio lever press

3	2	1	1	2	3	4	5	6
4	4	6	7(2)	5(1)	7(1)	9(2)	9(2)	11(1)
3	4	6	5(2)	5(1)	8(3)	8(1)	9(2)	10(0)
3	2	6	4(1)	6(3)	5(1)	9(2)	10(1)	10(1)
4	6	4	6(2)	6(1)	5(2)	7(2)	9(1)	11(1)
4	6	6	5(2)	6(3)	7(1)	10(2)	10(1)	11(0)
4	6	6	7(3)	7(1)	9(1)	10(2)	9(0)	11(1)
5	5	5	5(1)	5(2)	6(2)	4(1)	7(1)	9(2)
3	4	7	5(2)	8(3)	9(1)	10(1)	11(2)	10(1)
7	6	9	8(1)	6(1)	6(3)	9(1)	10(2)	10(1)
9	7	7	5(2)	7(2)	11(2)	10(0)	12(1)	12(2)
3	5	5	6(2)	4(1)	7(1)	7(3)	10(2)	
4	7	4	6(3)	5(1)	7(2)	9(2)	11(1)	
4	4	6	5(3)	7(2)	9(1)	9(2)	11(1)	
6	4	7	5(1)	6(2)	5(1)	7(2)	9(2)	
5	6	5	6(2)	4(1)	6(2)	8(2)	9(1)	
2	4	3	5(2)	6(2)	7(2)	8(2)	8(1)	
4	5	4	4(1)	6(2)	5(1)	7(3)	10(2)	
2	3	7	6(2)	6(2)	5(1)	8(3)	9(1)	
4	5	6	5(3)	8(1)	9(1)	10(2)	10(1)	
6	5	5	5(3)	6(2)	10(2)	10(1)	10(1)	
4	5	4	6(4)	5(1)	7(2)	9(2)		
4	5	6	6(3)	7(2)	8(2)	9(2)		
6	4	6	5(3)	6(2)	7(2)	9(2)		
4	5	4	5(3)	6(1)	7(2)	8(2)		
4	5	4	5(3)	5(2)	4(2)	8(1)		
2	3	5	6(3)	6(2)	8(3)	8(1)		
3	5	4	6(2)	6(2)	9(2)	9(3)		
3	5	6	5(2)	5(3)	7(2)	8(2)		
3	5	6	4(3)	9(2)	9(2)	8(2)		
2	3	6	6(2)	5(3)	7(1)	6(2)		

