the interictal EEG's of patients with epilepsy (8). Ingenious computer programs have been developed for detection of subcortical spike activity by analysis of surface EEG's coincident with subcortical transients (9). By coupling a reaction-time test to examination of brief samples of EEG spectra averaged just before stimuli that precede prolonged or faulty responses, we have found a pattern that resembles power-frequency spectra coincident with known spike activity and is mathematically consistent with such an interpretation. Demonstration of such spectra before prolonged responses from patients with generalized epilepsy confirms evidence that a deep spike focus is not uncommon in this disorder (10). If psychologic tasks are developed which permit better discrimination of pathological interruptions in the thought stream, then the ramp spectra preceding delayed or erroneous response may be of some practical use for localizing subcortical abnormality without implantation of intracranial electrodes.

J. R. STEVENS*
B. LONSBURY
S. GOEL
Division of Neurology, University of Oregon School of Medicine, Portland 97201

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* Address (1972–73): Department of Psychiatry, Massachusetts General Hospital, Boston 02114.

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Sexual Behavior: Ultrasonic Postejaculatory Song of the Male Rat

Abstract. *During the refractory period that follows ejaculation, the male rat regularly emits 22-kilohertz vocalizations. These cease after about three-fourths of the total period has elapsed, and this corresponds to an "absolute refractory period" during which the male cannot spontaneously initiate copulation. Similar 22-kilohertz vocalizations occur in other social contexts, and in general they appear to be desist-contact signals.*

Rats, mice, and many other rodents emit ultrasonic vocalizations (*1, 2*). The ultrasonic cries of newborns have received the greatest attention, but adults too produce and hear (*3*) these sounds. The vocal repertoire for social communication of the adult rat, *Rattus norvegicus*, consists primarily of ultrasonic calls. Sewell has described three such vocalizations: a 22-khz call emitted by defeated or supine subordinate males, a 50-khz pulse associated with aggressive activity, and a 50-khz vocalization that occurs during mounting activity (*2*).

The sexual behavior of the male rat has been exhaustively studied (*4, 5*). In brief, a series of mount bouts that culminate in intromission (penile insertion) leads to ejaculation, a postejaculatory refractory period follows, and a new series begins. This pattern is repeated for several ejaculations and after each, the postejaculatory interval (PEI) grows longer. The female generally does not hop, dart, and ear-wiggle during the PEI, whereas these behaviors are frequent in the copulatory series leading to ejaculation (*6*). The change in the behavior of the females toward refractory males may be influenced by the ultrasonic postejaculatory "song" of the male, the subject of this report.

This call was discovered during a standard observation of sexual behavior, during which ultrasounds were monitored with a Holgate ultrasonic receiver (bat detector) (*7*). Shortly after ejaculation, long, regular pulses were heard from the receiver, which was tuned to about 22 khz. These emissions were also viewed on an oscilloscope and appeared to be quite pure tones of frequency comparable to that indicated by the receiver. The female was removed from the observation chamber and the sound persisted. Later we observed that the calls could be correlated with long exhalations by the languid, refractory male rats.

The call was subsequently observed and recorded from a number of male Long-Evans rats of different ages and experience. Five sexually experienced males (approximately 150 days old), three old breeder males (12 to 18 months old), and three sexually naive male rats (70 to 90 days old) were subjects. Each was observed during at least two mating tests at intervals of approximately a week. Tests were carried out until either four PEI's had been recorded or spontaneous cessation of mating for more than 30 minutes occurred.

Female rats used as stimuli were prepared with injections of estrogen and progesterone (*8*). They were placed into the testing chamber, a 10-gal aquarium (40 by 26 by 29 cm) with cedar shavings on the floor, 5 minutes after introduction of the male; sexual responses—mounts with pelvic thrusting, intromission, and ejaculation—were recorded on a pushbutton-actuated strip chart recorded. Emission of the 22-khz call was monitored both auditorily with the bat detector and visually on an oscilloscope. The overall duration of calling was recorded on another channel of the chart recorder. The criterion for song production was the visible 22-khz signal on the oscilloscope screen.

Ultimately, the vocalization was recorded and characterized (*9*). It was found to consist of pulses of 22 to 23 khz, the duration of which ranged between 1 and 3 seconds. These pulses, emitted about a 0.25-second apart, formed trains of variable lengths (Fig. 1). Amplitude changes were apparent within and between pulses. At times the intensity of the sound was as high as 80 db (referenced to 0.0002 dyne/cm^2) measured 5 to 10 cm from the rat's head.

Each of the 11 males tested was observed to emit the vocalization during the PEI. None was observed to produce it during any other phase of the sexual sequence. There was considerable variation in the quantity and patterning of the calling. Some males called little during the PEI and others called almost continuously for the first three-fourths of the refractory period. Figure 2 shows the periods of vocalization for all of the animals over three PEI's. Calls generally began shortly after ejaculation

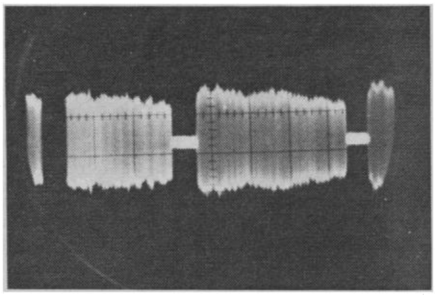

Fig. 1. Oscilloscope trace of a segment of a train of song pulses. Sweep speed was 0.5 sec/cm (1 cm = side of grid). Input to the oscilloscope was from a tape recording of the vocalization (*8*).

and faded after about three-fourths of the total period. The ratio of time of song termination to PEI was 0.75 ± .034 (mean ± standard error of the mean) for PEI 1, 0.74 ± .038 for PEI 2, and 0.66 ± .049 for PEI 3.

During much of the time that calling was recorded, the males lay quietly on the substratum, but at times calling was observed when the subjects were grooming or moving slowly about. The vocalization was disorganized by movements of the paws over the nose and mouth, but it did not cease. We cannot state whether the sound is emitted through the nose or mouth, but it is clearly correlated with exhalation movements 1 to 3 seconds long. The females tended to stay away from the males during the PEI and were rarely seen to hop or dart as they commonly do while the male is active. At times females approached the males and groomed them, or sat nearby.

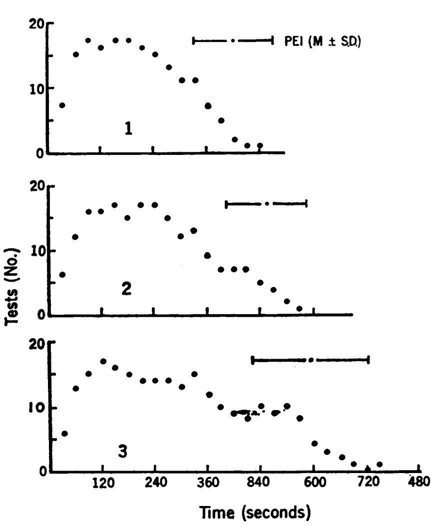

Fig. 2. Frequency distribution of song occurrence in terms of the number of tests in which vocalizations were observed during successive half-minute intervals of the PEI. Results for PEI's 1, 2, and 3 are given; there were 19, 20, and 20 tests, respectively, for these PEI's. Eleven subjects are represented in each case, each for no more than two tests. The PEI, mean (M) ± standard deviation (S.D.), is shown for reference in each graph.

Precisely what, if anything, the postejaculatory call of the male communicates to the female (or to other males) is not established, but it appears that the call reflects a state of social withdrawal; it could be a desist-contact signal. This interpretation is substantiated by other observations of vocalizations at the same fundamental frequency. We have observed the 22-khz subordination call (*10*) and find it quite similar to the postejaculatory call that we describe here. We have also observed 22-khz vocalizations to be emitted by stimulus females when they resisted the mounts of overly attentive males. Additionally, calls at the same frequency have been recorded from solitary male rats placed in a cage following rough handling or after receiving a brief (0.5-second) 1-ma shock to the skin. Perhaps 22 khz is a basic "carrier frequency" for signals denoting states of contact avoidance.

The PEI has been suggested to have relative and absolute phases (*11*). The absolute refractory period was thought to be that during which ro spontaneous copulatory activity can begin. Exogenous arousing stimuli, however, such as handling, can reduce the PEI, but even as insistent a stimulus as electrical shock did not decrease it to less than 75 percent of its control value (*12*). As judged from the shock studies, the absolute refractory period must occupy about 75 percent of the PEI (*13*), and this is strikingly similar to the mean percentage of the total PEI during which song occurs. It is suggested that the postejaculatory song functions to inhibit the copulatory behavior of the female during the period when the male is incapable of sexual responses, and that the vocalization is an auditory concomitant of the absolute refractory period.

The PEI reflects a state of behavioral inhibition (*4, 14*); electroencephalographic recordings during this period show a predominance of sleeplike activity (*15*). Similarly, movement is suppressed in other cases where 22-khz vocalizations are emitted. It seems likely that there is a great deal in common, physiologically, between the several states in which 22-khz vocalizations are emitted. Whether the 22-khz calls observed in different behavioral situations are the same or related vocalizations and whether they reflect similar physiological states cannot be stated. Further analysis of the several calls and of the conditions under which they are produced should supply these answers.

RONALD J. BARFIELD
LYNETTE A. GEYER

Department of Biology, Livingston College, Rutgers University, New Brunswick, New Jersey 08903

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7. This instrument is a heterodyne receiver similar to that described → J. D. Pye and M. Flinn [*Ultrasonics* **2**, 23 (1964)]. It is manufactured by Holgate's of Totton, Southampton, SO4 3ZF, England.
8. Ovariectomized females received 30 μg of estradiol benzoate and 500 μg of progesterone 54 and 6 hours, respectively, before the time of mating tests.
9. The vocalization was picked up by a Bruel and Kjaer 0.64-cm microphone, model 4136, with cathode follower. The signal was amplified and recorded on magnetic tape at 95 cm/sec on a Precision Instruments 6200 tape recorder. The tape-recorded signal was played back at 1/10 this speed and analyzed on a sound spectrograph, Kay Sona-Graph, model 7029A.
10. Sewell first described this vocalization (*2*). In a remarkable observation, Seward noted that male rats that had been defeated showed a "marked arhythmia" in their breathing. He was, of course, observing the motor concomitant of the subordination call [J. P. Seward, *J. Comp. Psychol.* **38**, 175 (1945)].
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