

The squeal as an indicator of aggression in mice*

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Fighting behavior of paired male mice was observed while automatic recording equipment counted the squeals. A strong positive relationship was found between the squeals and other measures of fighting. The results are discussed in terms of the validity of squeals as a measure of aggression and the usefulness of the squeal measure and the advantages it provides, i.e., the possibility of continuous monitoring of fighting behavior, elimination of O effects, less handling of Ss, and use of a "home cage" type of environment.

In the past, numerous techniques and dependent measures have been employed to assess aggressive behavior in mice and rats. Notation of specific behaviors was used by Beeman (1947) who observed mice in round robin fighting and devised a rating scale based upon the behavior observed. Other types of rating scales have been devised and used to measure aggression; however, these scales have usually been conducted with rats (see Davis, 1933; Yerkes, 1913; and Utsurikawa, 1917).

Latency to fighting has been used by several investigators (Fredericson, 1949, 1952; Fredericson, Fink, & Parker, 1955), while the actual number of attacks was used by Brown (1953) and Catlett (1961). Catlett also concluded that latency to attack, number of attacks, and accumulated attack time were all effective and accurate measures of aggression.

The amount of physical injury has also been used as a measure of aggression. Brown (1953) counted the number of wounds, and Fox and Snyder (1969), who checked the cages for mice who were either dead or dying of wounds, also counted the number of new bite marks on the Ss by postmortem examination. The number of deaths has been used by Crew & Mirskaia (1931) and Retzlaff (1938) as a dependent variable.

Thor, Hoats, and Thor (1970) and Thor and Hoats (1970) have measured aggression in rats by counting the number of loud vocalizations. Although the relation between squealing and fighting in mice has been frequently mentioned, it has not been systematically explored. Hall (1941) listed the squeal as a reaction assumed to be aggressive in mice and rats, and Scott (1966) notes that when a mouse is beaten, it squeals and runs away.

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Kahn (1951) mentioned squealing in his research on severe defeat, and Scott (1946) found that the pain caused by a tail pinch elicited squeals and escape attempts from young mice. The purpose of this study was to explore systematically the relation between squealing and fighting.

METHOD

Subjects

The Ss were 30 random-bred male Swiss Webster white mice approximately 4½ months old at the beginning of the experiment. The Ss had been individually housed for 2 months preceding the study and were housed individually during the experiment. Ad lib food and water conditions prevailed throughout the study.

Observers

Three Os recorded the fighting behavior of the paired mice. In addition to the senior author, two undergraduate students participated as Os. The two students received training

in the observation and recording of mouse fights but were not informed of the hypothesis or of the way in which the data would be handled.

Apparatus

The apparatus consisted of a mouse squeal detection circuit (see Morgret, 1972), a Grason-Stadler soundproof box, a cumulative recorder, and counters. The soundproof box was modified by the addition of a 16 x 8 in. Plexiglas window. The "event" channel of the cumulative recorder and a Hunter KlocCounter were operated by a hand switch held by the O and were used to record the duration of a fight. This switch also operated a counter to record the number of individual attacks. The mouse squeal detectors automatically operated a digital counter and the reinforcement marker on the cumulative recorder so that fighting and squeals could be recorded concurrently and according to their time sequence (see Fig. 1 for sample data tape). The cumulative recorder and counters were enclosed in an ice chest to silence the mechanical sounds associated with squeal recordings.

Fight trials were run in an 18 x 6 x 6½ in. plastic rat cage. A 15-W lightbulb above the cage provided illumination.

Procedure

Each two Ss, housed individually in a divided cage, were placed into the experimental chamber, and the recording equipment was activated. The O then watched the Ss for a period of 20 min and pressed the hand-held switch whenever a fight occurred and held the switch down for

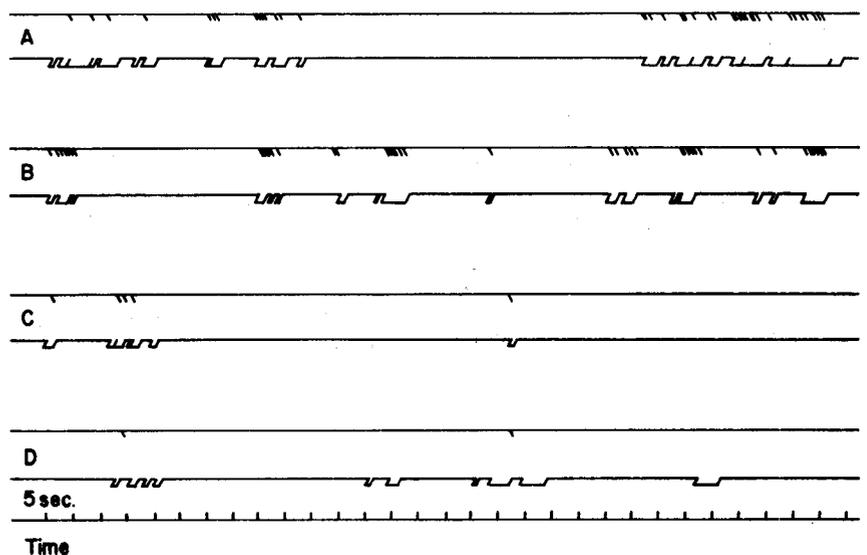


Fig. 1. Temporal relationships between squeals and fighting. Upper line shows the occurrence of a squeal as automatically recorded and the lower line shows the periods of fighting as measured by O.

as long as the fight lasted. At the end of the 20-min period, the Ss were returned to the home cage, the fight chamber was cleaned with Lysol, and clean litter was added. All pairs were tested for 12 observation periods, for a total observation time of 4 h per pair.

RESULTS

Observations

Since fights occur as closely spaced groups of individual attacks, a fight was defined as including any number of separate attacks; and the end of the fight was defined as the termination of an attack which was followed by a period of at least 15 sec without any aggressive encounters. Figure 1-A shows two fights, each one consisting of several attacks. Any attack occurring after the 15-sec nonfight period was considered a separate fight.

The sample tapes show other phenomena which were observed, although 1-A is most typical. Figure 1-B shows fights in which the individual attacks were less tightly grouped, while 1-C shows the close relation between fights and squeals. Note especially in 1-C the one isolated attack with its associated squeal. Even very short fights frequently had squeals associated with them. Figure 1-D shows two fights which occurred with only one squeal associated with each fight, and it also shows a fight without a squeal. While these latter two events were infrequent, they did occasionally occur.

Correlations

The data were first analyzed in terms of the correlation between number of squeals and other fighting behaviors, specifically: time fighting and number of fights. Each three successive 20-min periods were combined to form 1 h observation, and the correlations were calculated between the number of squeals and the time fighting for four 1-h periods. The range of these correlations extended from .48 to .98, with the mean correlations being .76, which accounts for over half of the variance (58%). Correlations were also calculated between the number of squeals and the number of individual attacks on an hourly basis. The range of correlations extended from .25 to .98, with the mean $r = .72$, which accounts for 52% of the variance.

The data were also examined in terms of conditional probabilities. Given a squeal, the probability of a fight was quite high (.83 overall), while the probability of recording a squeal during nonfighting behavior was quite low (.03 overall). These conditional probabilities confirm the relationship shown by the correlations.

DISCUSSION

The results indicate that the squeal can be used as an indicator of agonistic behavior. The relation between squeals and fighting is neither perfect nor necessarily direct; however, the relation is generally quite good. While this particular measure may not replace previous measures of aggression, it can make an important contribution, primarily in terms of using squeals to study aggression on a round-the-clock basis.

In observations of this type, the question of E effect must be taken into consideration. Since the apparatus is objective and therefore providing consistent data, the variances noted must be contributed by the Os. (O means for squeal/time fighting, squeal/attacks, and conditional probabilities were: O A—.97, .68, and .78; O B—.61, .75, and .82; O C—.70, .84, and .88.) The two student Os were not given knowledge of the hypothesis until after the observations had been completed; only one O was fully aware of the hypothesis under test. Therefore, an E effect in support of the hypothesis can be ruled out, since the only O who was aware of the hypothesis yielded the lowest conditional probability and the lowest r between squealing and number of fights. These O differences are not in agreement with Scott and Fredericson's (1951) statement that "True fighting behavior is unmistakable ... thus, there is no question of validity and in most cases, very little question of reliability ... [p. 277]." There seems to be some question of O differences since the Os in the present study received training, and during that time, agreed that the presence or absence of fighting did appear unmistakable. Still, the correlations and conditional probabilities varied from O to O. One possible explanation for this variability is that being unable to hear the squeals, the current Os could not use that cue to define a fight as previous Os could.

It should be noted that the greatest amount of variability was observed in the correlation between the number of squeals and number of fights. During a fight, the Ss frequently move apart for what may be either the end of the fight, or they may be separated for only a brief moment whereupon they resume active fighting. The scoring of this break in the moment when the Ss are moving apart calls for a very quick decision and could be scored as two separate attacks or one longer attack period. This appears to be a prime possibility for scoring difference. Whatever the source of variability, the use of the presently available automatic equipment would eliminate

this variability. Furthermore, if the O variability were eliminated, the relationship between squeals and fighting would most likely be higher than the observed correlations indicate.

The proposed procedure gives several other advantages over previously used aggression assessment techniques. In addition to ruling out the problem of O effects, the automatic measurement of squeals is also advantageous because handling effects can be minimized. The Ss' environment can remain relatively constant since they live in a "home cage" type of environment and are disturbed only for feeding, watering, and cleaning of the cages. It is possible, by using several large water bottles and a large amount of food, to leave the Ss completely undisturbed for several days.

Perhaps the greatest advantage of using the squeal as a measure of aggressive behavior is that it allows for continuous measurement of fighting. Other measures are limited in time and are therefore open to question since much data can be missed (see Ulrich, 1938). Data which were unavailable with past measures are now available to perhaps shed new light on mouse aggression.

It should perhaps be noted that squeals may not be a direct measure of aggression. That is, the current procedures do not permit separation of squeals emitted by the aggressing and those emitted by the hurt mouse. Previous research (Scott, 1946) indicates that squeals occur when a mouse is being hurt, which would suggest that squealing is a measure not of the agonistic behavior itself but of the effects of this behavior on the other mouse.

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from the pattern generator. The pattern is composed of two frequencies given in either an ABA or a BAB sequence, selectable remotely. The ON times for the t_1 , t_3 , and t_5 tone bursts are individually adjustable from 0.1 to 1.0 sec. The OFF times, t_2 and t_4 , are also adjustable within the same range as described above. The intercycle interval, t_6 , was designed to be adjustable from 0.2 to 2.0 sec. The two pure tones are generated by two oscillators not included in the pattern generator.

Figure 2 shows a schematic of the circuit design for the device. The individual ON and OFF durations, t_1 through t_6 are produced by a closed loop composed of six one-shot multivibrators, labeled OS₁ through OS₆ in the schematic. Commercially available circuit modules (Type EM 5011, Electronic Modules Corp., Timonium, Md.), chosen for their ease of mounting and small size, were incorporated in the device, although any other one-shot multivibrator having identical characteristics (e.g., OS-204 BRS-Foringer, Beltsville, Md.) can be used successfully. The inset in Fig. 2 labeled A shows the connections around the one-shot and gives the values of the RC network required to provide the necessary dwell times.

The design of the generator employs diode-relay logic throughout its construction. All relays used are ordinary 24-V types with the exception of those used in conjunction with the switching of pure tones (RD₁ and RD₂). These relays are of the 6-V variety and have mercury-wetted contacts to insure against switching transients. Relays RY₁ and RY₂ are spike-suppressed by Diodes D₄ and D₅, respectively. Any diode with a minimum breakdown voltage of 200 V is satisfactory. The requirements for Diodes D₁, D₂, and D₃ can be less stringent than those for D₄ and D₅. The C₁, a 50-microF 50-V capacitor provides a time delay for RY₁, which helps insure a chatter-free switching of

An inexpensive pure-tone pattern generator

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A low-cost sound pattern generator that is remotely programmable has been developed to produce pure-tone auditory patterns. The device, which is compatible with relay programming equipment, can generate either of two auditory patterns in the sequence ABA or BAB. The generator features an independently settable range of ON and OFF durations (0.1 to 1.0 sec) for pure-tone bursts and a selectable range (0.2 to 2.0 sec) of intercycle intervals. The inexpensive construction cost coupled with the flexibility of the timing characteristics of the generator should make it of interest to a wide variety of auditory researchers.

This report describes a low-cost remotely programmable sound pattern

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generator developed to investigate auditory pattern discrimination in pigeons¹ (Krasnegor, 1971). Because the device is so inexpensive to construct and has a flexible range of ON and OFF times, it should be of wide interest to auditory researchers. Figure 1 depicts two full output cycles

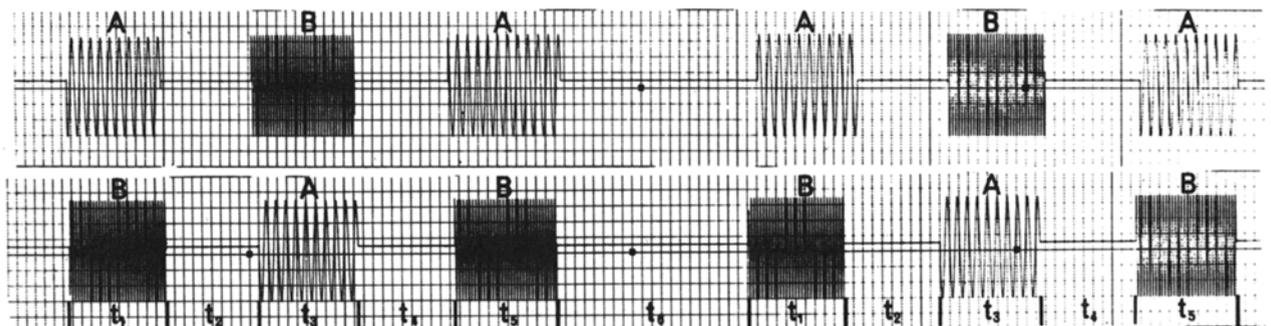


Fig. 1. Typical pattern generator output. The tracing shows Pattern ABA above and Pattern BAB below. In this example, the ON and OFF durations are 1 sec in length; intercycle interval duration is 2 sec.