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The Effect of Amygdalectomy on Orienting and Classical Conditioning in Monkeys

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SIMPLE REPETITION OF an experience modifies the behavior of organisms in predictable ways. Initially, an orienting reaction is obtained; this habituates. Any change in the repetition results in dishabituation. The proper coupling of the repetition with a reinforcer such as food or shock overrides habituation. The repeated omission of the reinforcer results in extinction of the behavioral pattern.

Over the past decade, our laboratory has been engaged in experiments designed to discover the role of one part of the brain, the amygdala, in this process of behavior modification. The amygdala was chosen because of the dramatic and easily observable effects of its removal (hyperphagia, "tameness") and because some other types of behavior modification—*e.g.*, simple sensory discrimination learning—are little affected by the lesion (Pribram and Bagshaw, 1953).

The initial experiments showed that amygdalectomy virtually eliminated the viscero-autonomic responses, including the galvanic skin response (GSR), cardiac acceleration, and respiratory disruption, which are normal components of orientation to a novel stimulus (Bagshaw, Kimble and Pribram, 1965). Behavioral components of orientation such as increased general activity and response to cue change were not disrupted (Schwartzbaum, Wilson and Morrissette, 1961; Schwartzbaum and Pribram, 1960; Douglas, 1966). No evidence of classical conditioning of the GSR to the offset of a light using a shock reinforcer was noted in a subsequent study (Bagshaw and Coppock, 1968).

Two possible formulations would account for these results: (1) amygdalectomy could alter viscero-autonomic reactivity alone, possibly raising the sensitivity threshold to the stimulus, or (2) amygdalectomy might impair the ability of all response mechanisms involved in orienting, habituation, and conditioning to condition.

A partial answer to the question of sensitivity threshold was obtained by studying the GSR threshold to shock. Amygdalectomy was shown to produce a slight but significant lowering of sensitivity threshold (Bagshaw and Pribram, 1968). All viscero-autonomic responses, but neither the ear flick nor some aspects of the EEG, were altered by amygdalectomy. Still, certain deficiencies in these studies left some doubt as to the results. The orienting-habituating studies used only the onset of tone, a very mild stimulus. The conditioning study neglected to use adequate pseudoconditioning and sensitization controls, and measured only viscero-autonomic responses.

Thus, the experiments reported in this article were undertaken to provide more definitive data—for the answers to these questions are important. If orienting and classical conditioning are always disrupted by amygdalectomy, then these behavioral processes acquire a special biologic status by virtue of their relation to this specific part of the brain. If only the viscero-autonomic aspect of the orienting and conditioning mechanism but not the viscero-autonomic mechanism itself is affected, which is most likely, the question is raised of the import of viscero-autonomic activity in behavior modification.

Experiment 1 repeats the earlier Bagshaw and Coppock study (1968), but tests orienting, habituation, and classical conditioning responses to an intense, unconditional stimulus, including an assessment of heart rate and respiratory rhythm in addition to the GSR. Experiment 2 uses

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a skeletal (temporal muscle) response as an indicator of orienting and conditioning. Both studies control for possible pseudoconditioning effects.

Experiment 1*

Method

Subjects. A total of 17 adult rhesus monkeys (*Macaca mulatta*) weighing between 3.8 and 4.9 kg were used. Seven of these had been subjected to bilateral amygdalectomy (Group AM). The surgical procedure has been described previously (Bagshaw, Kimble, and Pribram, 1965) and is briefly noted in Experiment 2. Of the unoperated normal animals (Group N), 7 were given the paired paradigm, three were used as unpaired controls. All subjects were naive and had not previously been used for behavioral testing. They were housed in individual cages in a group laboratory facility and all were fed a normal laboratory diet.

Histologic verification of the lesions of the operated subjects may be found in Bagshaw and Pribram (1972). In general, the resections were complete (total) with minimal damage to surrounding structures.

Apparatus. A Grass polygraph, Model V-D, was used to record electrocardiographic (EKG) respiratory excursions, and skin resistance was measured on a Fels Dermohmeter. Respiration was measured by means of a pneumographic tube placed around the animal's abdomen and connected to a PT-5 pressure transducer that converted the volume changes to voltage changes. Pure tones were generated by a Grayson/Stadler, 905D twin oscillator, and were presented through a speaker positioned 70 cm above the vertex of the animal's head. A constant background of white noise was provided by a small rotary fan. Light was generated by a 10 w red bulb located above the animal's head, near the speaker. EKG and respiratory data were amplified through Tektronix amplifiers, and a laboratory-built 10 × amplifier was used for the GSR signal. These three channels were fed on-line into a laboratory PDP-8 computer where the data were digitized and stored on magnetic tape for future analysis. The actual experiments were run on-line by the computer. The operator could use a manual start-and-stop button to interrupt the experiment if necessary. Signals were monitored on a Tektronix oscilloscope at all times. Shock was administered from a laboratory-built shock generator.

Procedure. The monkey was restrained in a Foringer primate chair. His ankles and wrists were comfortably secured to the chair to prevent his tampering with the electrodes. His lower legs and midchest region were shaved and scrubbed, first with Phisohex and water, then with alcohol to provide a clean surface for good electrode contact. EKG electrodes, consisting of small, curved, suture needles soldered onto pick-up leads, were placed subcutaneously on the chest, on either side of the heart, and across the sternum, and were taped in place. Subcutaneous placement eliminates movement and respiratory excursion artifacts. One Beckman GSR electrode was applied to both the plantar surface of the right foot and to the right calf. These electrodes were filled with Beckman electrode jelly and then secured in place with gauze bandages and Elastoplast wrappings.

Respiration was monitored with a pneumographic tube placed around the animal's abdomen. Shock electrodes were two metal EEG electrodes placed about 2 cm apart on the plantar surface of the left foot. Grass electrode paste was applied to the electrodes and to the skin to ensure good contact. The electrodes were secured in place with gauze, adhesive, and Elastoplast wrappings. Figure 1 shows the animal in the restraining chair with electrodes in place.

The animal was then placed in a darkened, sound-insulated chamber (Acoustic Laboratories, 120 × 120 × 215 cm) to reduce ambient noise. The animal was allowed a 10 minute period of equilibration, after which five minutes of control recordings were made. No stimuli were applied during this time.

The computer was then adjusted to initiate the trail phase. The computer was programmed to operate relays controlling the onset and offset of the conditional stimulus, (CS) and the unconditional stimulus (UCS). Data were recorded both on the Grass Polygraph and on magnetic tape by the PDP-8 computer. Polygraph recordings were continuous, but the computer collected only 45 seconds of data during each trial. The entire procedure took four to five hours. After testing, the animal was fed and returned to his home cage.

Each animal was tested in one session of a paired or unpaired classical conditioning paradigm.

In the paired paradigm, the conditional stimulus consisted of a compound stimulus—a pure tone (1000 cps) and a red light (10 w)—presented simultaneously for 15 seconds. One-half second after the offset of the CS, the unconditional stimulus—a 5 amp shock—was applied for 200 msec to the animal's foot. This constituted one trial. The interval between trials varied ran-

* Represents work done by Dr. Margot McNeil in partial fulfillment of the requirements for the Ph.D.

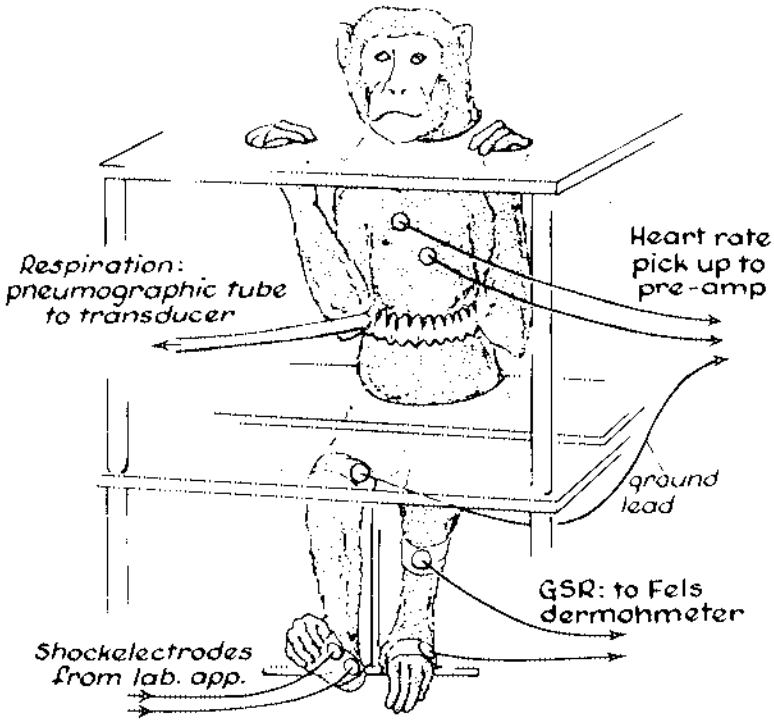

FIG. 1

domly from 45 seconds to 3.5 minutes, with an average duration of two minutes.

Fifteen "habituation" trials which consisted of the CS alone were first presented. Forty conditioning trials followed, consisting of paired presentations of the CS and the UCS as described in the previous paragraph. Finally, 10 extinction trials were given in which only the CS was presented.

In the unpaired paradigm, the procedure was generally the same as that for the paired paradigm except that the UCS (shock) was never presented in conjunction with the CS. Rather, the UCS followed it at some random interval which was never *less* than 30 seconds (post-CS offset) and at most, was 195 seconds. The number of trials was the same as that of the paired paradigm: 15 habituation trials of CS alone, 40 pseudoconditioning trials (CS and UCS but not in conjunction), and 10 extinction trials of CS alone.

Measures of heart rate. The EKG was recorded in digital measures of R-R intervals during the experiment. Heart rate data were analyzed by use of the PDP-8 computer. Measures of basal rate, orienting, and habituation were obtained before the following measures of heart rate were selected.

Basal rate. This measure was obtained during the initial resting period before presentation of the light and tone (CS). Five-beat samples were obtained one minute before testing and again from the last five beats before presentation of the CS. Mean *rates* of the two groups were calculated and compared.

Measurement of the orienting response (initial response to the CS). This measure was obtained from the first two trials only. The change in heart rate from its prestimulus baseline level, subsequent to the presentation of the CS, was taken as a measure of orienting. A total of 25 beats were analyzed; the mean of the intervals between the five prestimulus beats was the control baseline and was compared to the maximum interval between the 20 beats following onset of the CS. The mean was subtracted from the maximum to obtain the magnitude of the response.

Measurement of the conditioning response. This measure was obtained during the extinction trials (56–65) at the time when the shock would normally have been presented (0.5 seconds after the CS had been turned off). A total of 25 beats was analyzed. The maximal and minimal intervals between the 20 beats just after offset of the CS were compared with the mean of the intervals

between the five beats just preceding the offset of the CS.

Paired and unpaired groups of normal and amygdalectomized animals were compared using all measures.

Measures of GSR. Galvanic skin responses were scored when the response occurred between 0.8 and 5 seconds after onset of the CS. The decrease in resistance (ohms) from the onset to the peak of the response was measured, and any response of 300 ohms or more was accepted. Trials in which movement occurred in the control period just prior to onset of the CS were discarded. Because of the variability of the baseline rate of production of GSR in any one amygdalectomized subject over time (see autonomic lability below), the measure of GSR used was the ratio of the number of GSRs occurring just after onset of the CS on a trial, as described above, to the total number of GSRs occurring during that trial.

Measures of GSR were taken during the periods of: orienting in the first 10 trials, and conditioning during the 10 extinction trials (56–65), at the time just after offset of the CS when the shock normally would have occurred.

Measures of respiratory rate. Respiratory disruptions occurring during the first five seconds of the CS were rated on a scale of 0 to 3, with 0 equalling none observable, 1 equalling just noticeable, 2 equalling moderate, and 3 equalling marked. The control period was comprised of the five seconds immediately preceding onset of the CS, the end of the last intertrial interval. Irregularities as well as acceleration and deceleration were taken into account in the scoring. Measures of orienting and conditioning were obtained as was the case with the GSR.

Results

Cardiac Measures

Basal rate. Marked differences between the two groups (N and AM) appeared in the prestimulus control period before the testing was begun. The differences in heart rate between the AM and N groups was striking. Figure 2 illustrates the mean baseline rate for both groups in beats per minute (bpm). The AM group clearly had a much higher overall rate than that of the N group. The mean rates for the N group (202 bpm) and for the AM group (240 bpm) differ significantly ($p = .025$, Fishers + − test). Heart rates in the normal group ranged from 181 to 235 bpm, whereas the range in the AM group was from 204 to 288 bpm (Table 1). The latter rates are extremely high for rhesus monkeys. Such rates were never approached by the normal animals, even under conditions of pain or severe stress.

Over the course of the experiment, however, the amygdalectomized group did show a mean decrease of about 30 bpm by the end of testing so that their rates fell to within normal range.

Orienting. Significant differences were again observed between the N and AM groups in their orienting behavior. The normal animals, including the two normal subjects whose basal rates exceeded those of some of the amygdalectomized subjects, showed a strong cardiac acceleratory response to onset of the CS, whereas the AM group showed hardly any change in cardiac response—as if nothing had happened. The change in rate is significant for the normal subjects but not for the AM group ($p = 0.01$, Fisher's Test). Figure 2 shows the heart rate (in bpm) after the two initial presentations of the CS in comparison with the baseline rate for both groups (paired and unpaired stimuli combined) since there was no difference in conditions during habituation. Individual rates following onset of the CS may be found in Table 1.

The mean increase for the normal subjects was about 21 bpm; for the AM group it was 8 bpm, representing a 10% and a 3% change, respectively. The change scores themselves are significantly different between the groups ($p = .001$, Fisher's Test; $N = 10$, $A = 7$), even when corrected for differences in baseline activity. (The increase in rate for the AM group is attributable mostly to the response of one AM subject (AZ). This same subject showed the lowest initial heart rate of the AM group and is the one subject whose baseline rate was within the normal range.)

Conditioning. The period immediately following offset of the CS during extinction (the time at which the shock occurred during the preceding conditioning phase) was used to observe conditioning. Here, too, clear differences between the groups were noted. The results are tabulated in Table 2. The results are not statistically significant, but very definitive observations may be made. The AM group gave no reaction whatsoever to offset of the CS, thus showing *no* evidence of cardiac conditioning. The normal subjects, on the other hand, *did* show some type of response. Five of the 6 normal subjects showed a deceleration during the 20 beats following offset of the CS. Three of these subjects (N1, N2, and N3) showed a consistent deceleration over the entire period. Two subjects (N4 and N5) showed an initial deceleration followed by an acceleration to a level just above that recorded prior to offset of the CS. The remaining subject (N5) did not show a deceleration. Thus, most of the normal subjects

TABLE 1. Individual and Mean Baseline Heart Rates and Changes in Rate Upon Presentation (Onset) of Conditioned Stimulus (CS) Alone (Habituation)*

	Heart Rate (Beats per Minute)		
Subjects	*Baseline (Prior to 1st CS)*	*Orienting (Response to 1st two CS)*	*Change in Rate*
N1**	204.1	234.0	29.9
N2	181.4	194.8	13.4
N3	186.8	213.6	26.8
N4	192.6	219.8	27.2
N5	229.2	246.2	17.0
N6	207.1	223.1	16.0
N7	210.7	226.4	15.7
NU1	192.6	219.8	27.2
NU2	236.1	263.3	27.2
NU3	192.6	202.4	9.8
Mean	201.7	222.7	21.0
A1†	230.6	238.8	8.2
A2	203.7	227.3	23.6
A3	242.5	242.9	0.4
A4	248.7	252.5	3.8
A5	[213.5]	[217.2]	3.7
AU1	288.5	295.6	7.1
AU2	250.3	259.7	9.4
Mean	239.7	247.7	8.0

* Baselines are averages of five beats just prior to the onset of the first two stimuli; rates during CS are averages of five beats just following onset of first two stimuli. Data from all subjects are included since "paired" and "unpaired" groups do not differ in treatment during Habituation.

** N = normal subject

† A = amygdalectomized subject

showed some evidence of a cardiac conditioning response, whereas none of the AM subjects ever did. Their heart rates remained almost constant. The reactions of the unpaired control groups were similar to the amygdalectomized group; that is, they showed no change in heart rate in response to the offset of the CS.

GSR Measures

Orienting. Using the relative GSR as a measure, the normal animals were found to show a much higher percentage of orienting responses than the AM group. In the first 10 trials, the normal group responded at a level of 33% to the presentation of the CS, whereas the AM group scored only during the first 10 conditioning trials ($p = .05$, Median test). With further testing a decrease in the percentage response to onset of the CS occurred in the normal group; they habituated to the testing procedure. The scores of the two groups failed to show any significant differences in the remainder of the conditioning trials. Group scores for each of the 10 trial block periods are illustrated in Figure 3.

During the extinction trials, the percentage of response of the normal group to onset of the CS again increased while the rate of the AM group did not, so that the difference between the groups again became significant ($p = .05$, Median test). The normal subjects scored over 30%, whereas the AM group scored approximately 13%.

If this analysis is carried one step further, one finds that the amygdalectomized animals may be further subdivided into two distinct groups, both of which fall outside the normal range of activity.

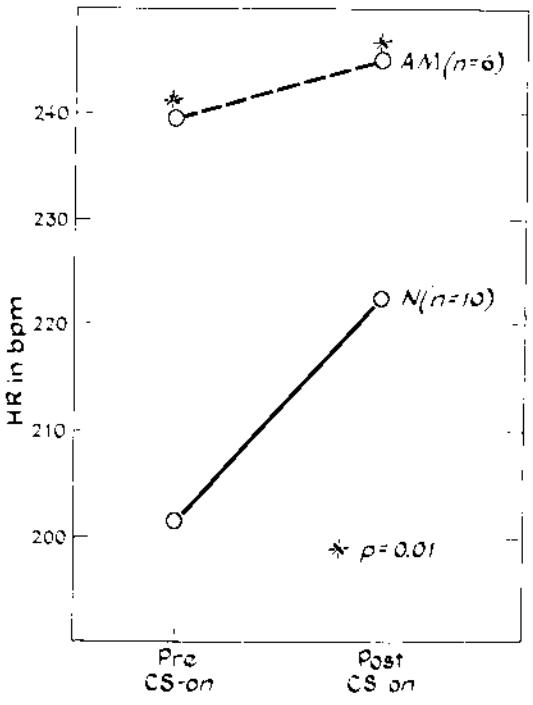

FIG. 2

Autonomic lability (background activity). When the total number of GSRs occurring during the intertrial time was used as a measure of spontaneous rate, the normal group (N = 11) showed a mean rate of 20 responses with a range of 18 to 22 responses. Not one amygdalectomized animal fell within this range! Their scores fell either well above or well below the limits of this distribution. They may be clearly divided into two groups: one whose mean response rate for this same period was 30 (range of 28 to 30); and another whose mean response rate was 7 (range of 2 to 11). These data correspond with the findings of Bagshaw, Kimble, and Pribram (1965) which revealed that amygdalectomized subjects either have very high GSR rates or produce virtually no GSR responses.

Conditioning. The primary measure of conditioning with respect to the GSR involves the relative responses of the N and AM groups to offset of the CS during habituation and extinction (*i.e.*, before and after conditioning). During habituation, the mean relative percentage of GSR to offset of the CS was 17.9% for normal subjects and 18.0% for the AM group. During extinction, the percentage rose to 33.5% for the N group and fell to 7.4% for the AM group, a significant difference ($p = .05$, Median test).

Respiratory Measures

Orienting. Measures of respiratory response to onset of the CS (disruption scores) failed to show any differences between the two groups during the initial trials. Both groups showed scores of normal, high respiratory disruption. In the first five habituation trials, group N averaged 2.5 (measured on a 3.0 scale), and group AM averaged 2.7.

During the final trials of habituation, the response of both groups was less than that noted during the first five trials. The data suggested that the amygdalectomized animals continued to respond to onset of the CS at a somewhat higher frequency and amplitude than did the normal subjects.

Conditioning. The normal animals showed a significant increase in the respiratory disruption score to offset of the CS, relative to the last five habituation trials (Fig. 4). This same phenomenon was noted in the normal group in the measurement of the GSR. In contrast to this, the AM group showed only a slight increase in responsivity. The difference between the groups was significant ($p = .05$, Median test). The response of the normal group to the paired and unpaired paradigms differed appreciably during extinction: the response of the AM group to the paired and unpaired paradigms did not.

Discussion of Experiment 1

The findings of this experiment may be summarized as follows.

(1) The amydalectomized subjects did not show the normal pattern of orientation—cardiac acceleration, increased frequency of galvanic skin responses, and respiratory disruption to the onset of a light and tone. Initially (before any stimuli), the amygdalectomized subjects had extremely rapid heart rates, and either extremely high or extremely low rates of GSR, *i.e.*, hyperlabile or hyperstable skin resistance. However, the lack of cardiac reaction of the amygdalectomized animals was not a result of a ceiling effect since normal subjects with similar high rates did show reactions. Furthermore, over the course of the experiment, the heart rates of the amygdalectomized subjects in response to onset of the CS decreased to the point where they were indistinguishable from that of the normal group. Their abnormal reaction to change or novelty was then manifested in their failure to reorient to onset of the CS when the UCS (shock) is discontinued during extinction.

TABLE 2. Individual and Mean Cardiac Responses to Offset of Conditioned Stimulus (CS) During Extinction Trials*

Subjects	Heart Rate (Beats per Minute)		
	Prior to CS Offset	Following CS Offset	Change
Paired			
N1**	212	192	−20
N2	233	216	−17
N3	135	122	−13
N4	166	154, 179	(−12)
N5	213	223, 210	(+10)
N6	218	212, 231	(−6)
Mean	196	(186)	(−10)
Unpaired			
NU1	213	212	−1
NU2	218	216	−2
NU3	225	228	+3
Mean	219	219	0
Paired			
A1†	238	238	0
A2	235	234	−1
A3	199	197	−2
A4	181	180	−1
A5	194	190	−4
Mean	209	208	−1
Unpaired			
AU1	192	190	−2
AU2	225	223	−2
Mean	208	206	−2

* Data include five beats prior to offset of CS and 20 beats following. Figures in parenthesis indicate means calculated the initial rate change of subjects N4, N5 and N6.

** N = normal subjects

† A = amygdalectomized subjects

(2) The amygdalectomized subjects did not show any conditioned viscero-autonomic responses to offset of the CS when this was paired with a shock UCS. Normal subjects revealed increased GSRs and respiratory disruption in response to offset of the CS during extinction as compared with their responses to the same offset of the CS prior to conditioning (*i.e.*, during habituation). They differ from the normal group who had never experienced paired presentations of CS and UCS. The unpaired subjects showed no difference in response to offset of the CS between habituation and extinction trials.

The normal subjects (5 of 6) showed a conditioned cardiac deceleration to offset of the CS, but in two of the five subjects, deceleration was followed by an acceleration above the pre-CS offset level within the 20 beats analyzed. Amygdalectomized subjects showed essentially no change in heart rate to offset of the CS, and hence showed no evidence of conditioning.

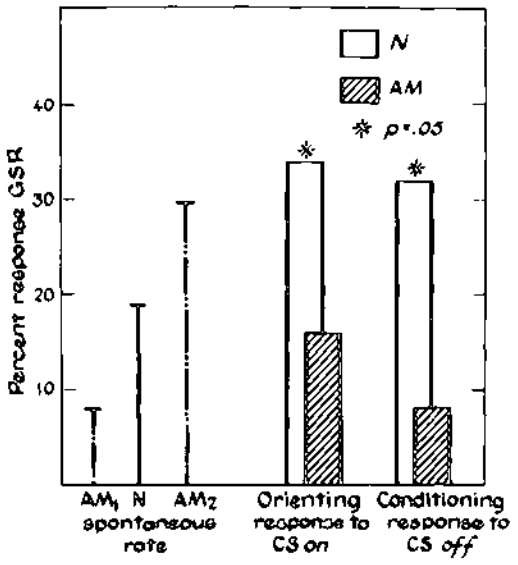

FIG. 3

Experiment 2*

Method

Subjects. The subjects were 6 unoperated and 6 bilaterally amygdalectomized immature rhesus monkeys (*M. mulatta*). Four subjects in each group had been used previously in a study in which their eye movements were monitored by line-of-site photography as they freely observed visual displays. The others had been trained with a series of visual discrimination reversals in an automated testing apparatus. All subjects had been used in Experiment 1.

The amygdala lesions had been made 2 to 3 years prior to the start of this experiment by subpial suction resection under direct vision via a transtemporal approach. The amygdala was removed bilaterally in a single operation.

Apparatus. During conditioning, the subjects were seated in a primate restraining chair; movement of their limbs was restricted to prevent displacement of the recording leads. The unconditional stimulus (UCS) was a 0.5 second train of pulses (1 msec²) generated by an AEL Laboratory Stimulator set at a sufficient intensity to reliably produce loud vocalization and movement in all subjects. The shock UCS was constant for all subjects and was administered to the second and index fingers through copper leads. The conditional stimulus (CS) was a 5.0 second light from a 100 watt tensor lamp located about two feet in front of the subject

Potential differences between two suture needles sewn into the temporal muscle and midline fascia of the skull were recorded on a two-channel Brush Chart Recorder and served as a conditional (CR) and unconditional (UCR) responses. Tektronix amplifiers were set to pass and amplify activity in the frequency range of 0.2 to 50 Hz, a range appropriate to record gross head and body movements, or to pass and amplify activity between 80 and 250 Hz, the EMG of the temporal muscle.

Presentation of the conditioning stimuli was controlled by a PDP-8 computer interface which also produced an event marker, recorded on a second channel of the Brush recorder.

Procedure. Three amygdalectomized and three unoperated subjects were assigned to a paired conditioning group and received the conditioning paradigm; the remaining 6 subjects were assigned to unpaired pseudoconditioning control groups. All four groups thus formed, received five consecutive training phases: 10 trials of habituation, 80 trials of conditioning, 20 trials of extinction, 35 trials of reconditioning, and 10 trials of re-extinction. During the conditioning and reconditioning phases, the paired group received trials in which the five second light CS was followed 0.5 seconds later by the 0.5 second shock UCS according to a two minute variable interval schedule. For the unpaired group, "conditioning" trials started randomly with either a CS or a UCS which was followed 30 seconds later by the unused event. Thus, the CS followed the UCS as often as it preceded it, with the constraint that the two stimuli never occurred less than 30 seconds apart.

The habituation and extinction phases were the same for both the paired and unpaired groups and consisted of presentations of the CS alone according to a two minute variable interval schedule.

Throughout all of the 155 trials, the amplified responses were continuously monitored. During the habituation period, the conditioning trials, and the first 10 extinction trials, gross movements were recorded through the low-pass filter from two of the three subjects in each of the four lesion-condition groups. The EMG was recorded through a high-pass filters in the four remaining subjects. During the remaining extinction trials and during the following reconditioning and re-extinction phases the filters were reversed so that EMG responses were recorded from subjects formerly providing movement responses, and movement responses were recorded from the subjects formerly providing EMG meas-

* Conducted while Drs. Reitz and Spevack were postdoctoral fellows.

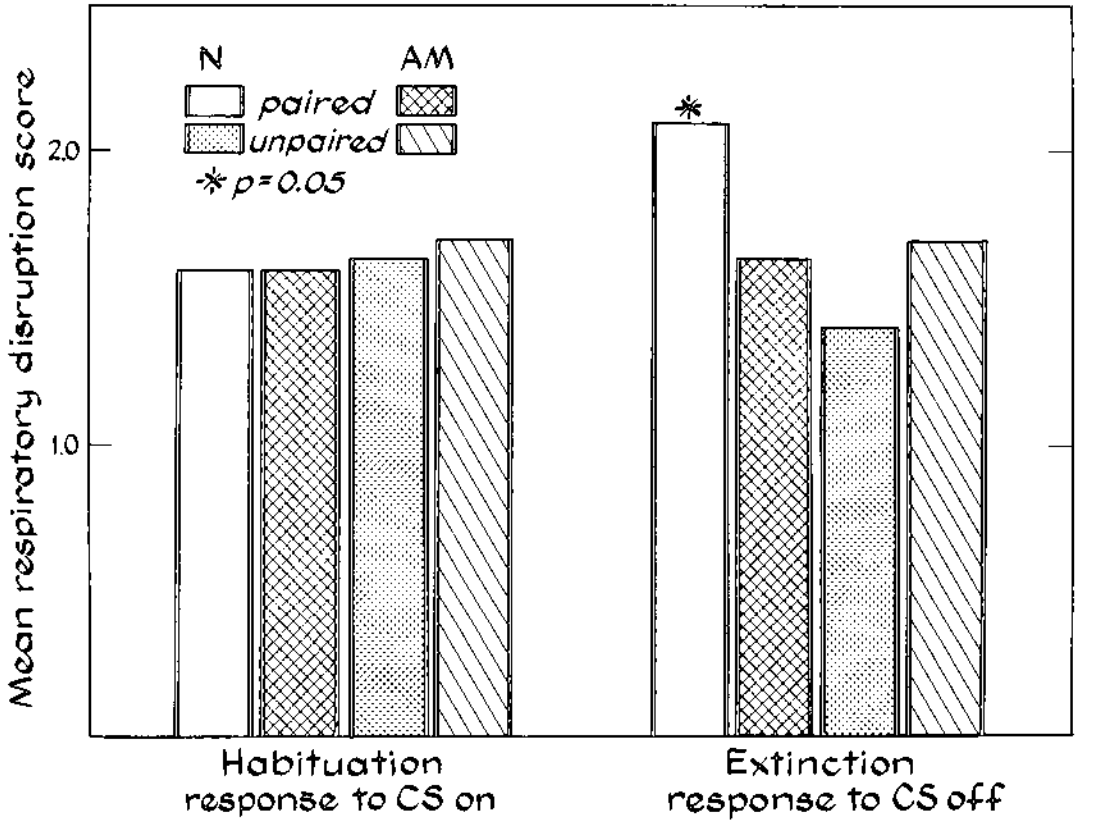

FIG. 4

urements. Both of these measures were used in order to examine possible CRs that might consist either of movements of the head and body or of a tensing of the temporal muscles.

Response measurement. The five seconds preceding the onset of the CS and the five seconds during which the CS was present were divided into 10 one second intervals (Vincentizing). The maximum excursion of the pen from baseline during each of these 10 intervals was measured: in this way, EMG or movement amplitude preceding and during the CS were represented in 10 scores. To generate a composite measure for each trial, the five scores for the CS period were summed and divided by the sum of the scores for all 10 intervals. If the resulting proportion was greater than 0.5, EMG or movement amplitude was greater during CS presentation than during the five seconds preceding the onset of the CS. Conversely, a proportion of less than 0.5 indicated that CS presentation reduced EMG or movement amplitude.

Results

General Observations

The proportions derived from movement recordings did not appear to differ from those derived from EMG records either in the rate at which they evidenced conditioning or in the final level of conditioning reached. Also, no obvious difference between the amygdalectomized and unoperated subjects was evident in this regard, although the small number of subjects prevented statistical verification of this conclusion. Lacking any obvious differences, however, the two response measures were combined for all subsequent analyses.

Amplitude Analysis

Figure 5 shows the mean amplitude of responses produced by the four lesion-condition groups during the five phases of the experiment.

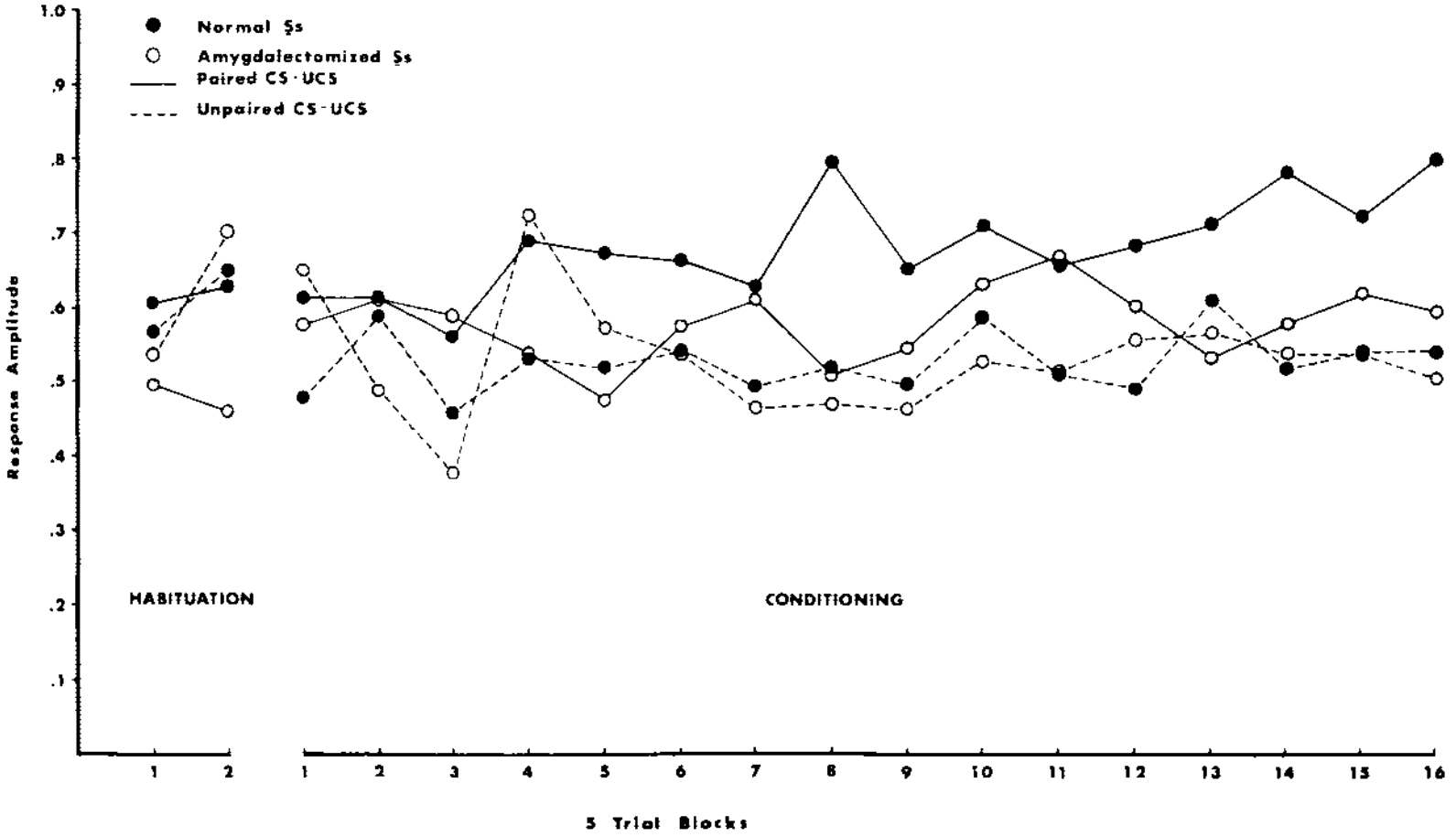
Normal Ss
Amygdalectomized Ss
Paired CS-UCS
Unpaired CS-UCS
Response Amplitude
1.0
.9
.8
.7
.6
.5
.4
.3
.2
.1
HABITUATION
CONDITIONING
1
2
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
5 Trial Blocks

FIG. 5A

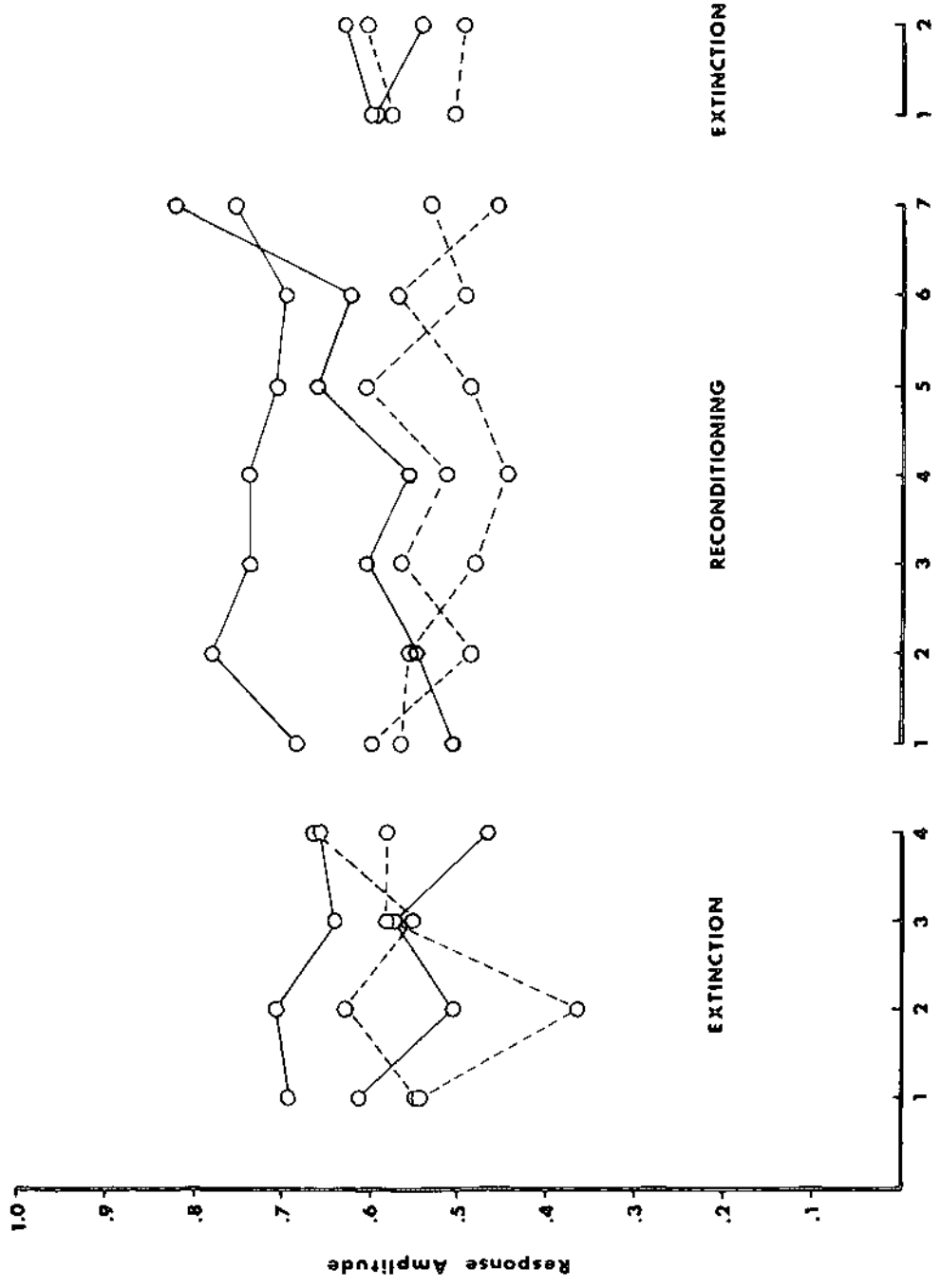
Response Amplitude
1.0
.9
.8
.7
.6
.5
.4
.3
.2
.1
EXTINCTION
RECONDITIONING
EXTINCTION
1 2 3 4
1 2 3 4 5 6 7
1 2
5 Trial Blocks

FIG. 5B

Despite extensive variability between the trial blocks, during the conditioning and reconditioning phases, the paired subjects produced larger amplitude responses to the CS than did the unpaired subjects. Moreover, during the same phases, the unoperated controls seemed to show responses of somewhat larger amplitude with onset of the CS than did the amygdalectomized subjects.

To statistically test these observations, a repeated measures test (Edwards, 1962), using the arc sine transforms of the proportions, was performed separately for each of the five phases of the experiment. These analyses assessed the differences in amplitude of EMG or movement between the lesion groups and the paired and unpaired groups over the trials comprising each phase of the experiment. Significant differences were apparent in the amplitude of response between the paired and unpaired conditions for the conditioning phases ($F = 17.76$; $df = 1/8$, $p \leq .01$) and the reconditioning phases ($F = 19.35$, $df = 1/8$, $p \leq .01$) of the experiment. No other differences were statistically significant for these or for the habituation and two extinction phases, including the trials effect. However, it is noteworthy that the differences in the amplitude of response between the amygdalectomized and intact subjects during the conditioning phases ($F = 4.57$, $df = 1/8$, $p \leq .07$) and the reconditioning phases ($F = 5.28$, $df = 1/8$, $p \leq .05$) is just beyond the .05 level of significance. (This suggested that the amygdalectomized subjects tended to produce conditional responses of smaller magnitude than did the intact subjects, even though they produced responses as frequently.) Rather than concluding that this was a result of the neurologic lesion, however, an alternative should be considered. Conditional responses were recorded from electrodes placed in the monkey's temporal muscle. This muscle had been sectioned during the surgical procedure for the amygdalectomized group. The tendency toward smaller conditional responses by the subjects with lesions may simply have been attributable to a difference in the size of the muscle.

In the analysis above, the amplitude of response was confounded with the frequency of response. Thus, the possibility existed that differences in amplitude and frequency of responding were distilled from the individual proportions; that is, a response was considered to have occurred if the proportion representing the relative EMG or movement change during the CS exceeded 0.5.

Frequency Analysis

Figure 6 shows the relative frequency of response by the four lesion-condition groups in five trial blocks during the five phases of the experiment. Substantially the same results appeared as were obtained when the amplitude of response was examined. Once again, much variability was apparent between the trial blocks, but differences in the frequency of response between the paired and unpaired groups and the lesion groups were again evident during the conditioning and reconditioning phases of the experiment. A repeated measures test was performed which assessed the differences in the relative frequency of response between the two training conditions and between the lesion groups over the five phases of the experiment. The number of trials in which EMG or movement proportions were observed to exceed 0.5 was expressed as a proportion of the total number of trials comprising each phase. Thus, the response probability of each subject was described by a single relative frequency score during each of the five phases of the experiment. The analysis was performed on the arc sine transforms of these relative frequency scores.

As seemed apparent from Figure 6, the paired subjects responded significantly more often during the experiment than did the pseudoconditioning control groups ($F = 7.17$, $df = 1/8$, $p \leq .05$). However, the amygdalectomized subjects paired and unpaired, did not differ in their frequency of response from the corresponding intact subjects (nonsignificant lesions and lesions × conditions interaction effects). The frequency of response did differ significantly in the five phases ($F = 6.44$, $df = 4/32$, $p \leq .05$), which can be attributed to differences between the paired and unpaired subjects in individual phases of the experiment (significant phases × conditions interaction: $F = 5.28$, $df = 4/32$, $p \leq .001$) and not to differences in the frequency of response between amygdalectomized and normal subjects across all phases (nonsignificant lesion × phases interaction). The triple interaction reached significance at the 0.01 level ($F = 4.03$, $df = 4/32$).

Orthogonal comparisons indicated that the frequency of response was significantly greater during the conditioning and reconditioning phases of the experiment than it was during the habituation and extinction phases. Also, significantly more responses occurred during the second extinction phase than during the first. No significant differences in the frequency of response were apparent between the conditioning and reconditioning phases.

Discussion of Experiment 2

The amygdalectomized and intact subjects who received punishment 0.5 seconds after each CS presentation (paired groups) showed significantly larger and more frequent responses than did those for whom shock was not contingent

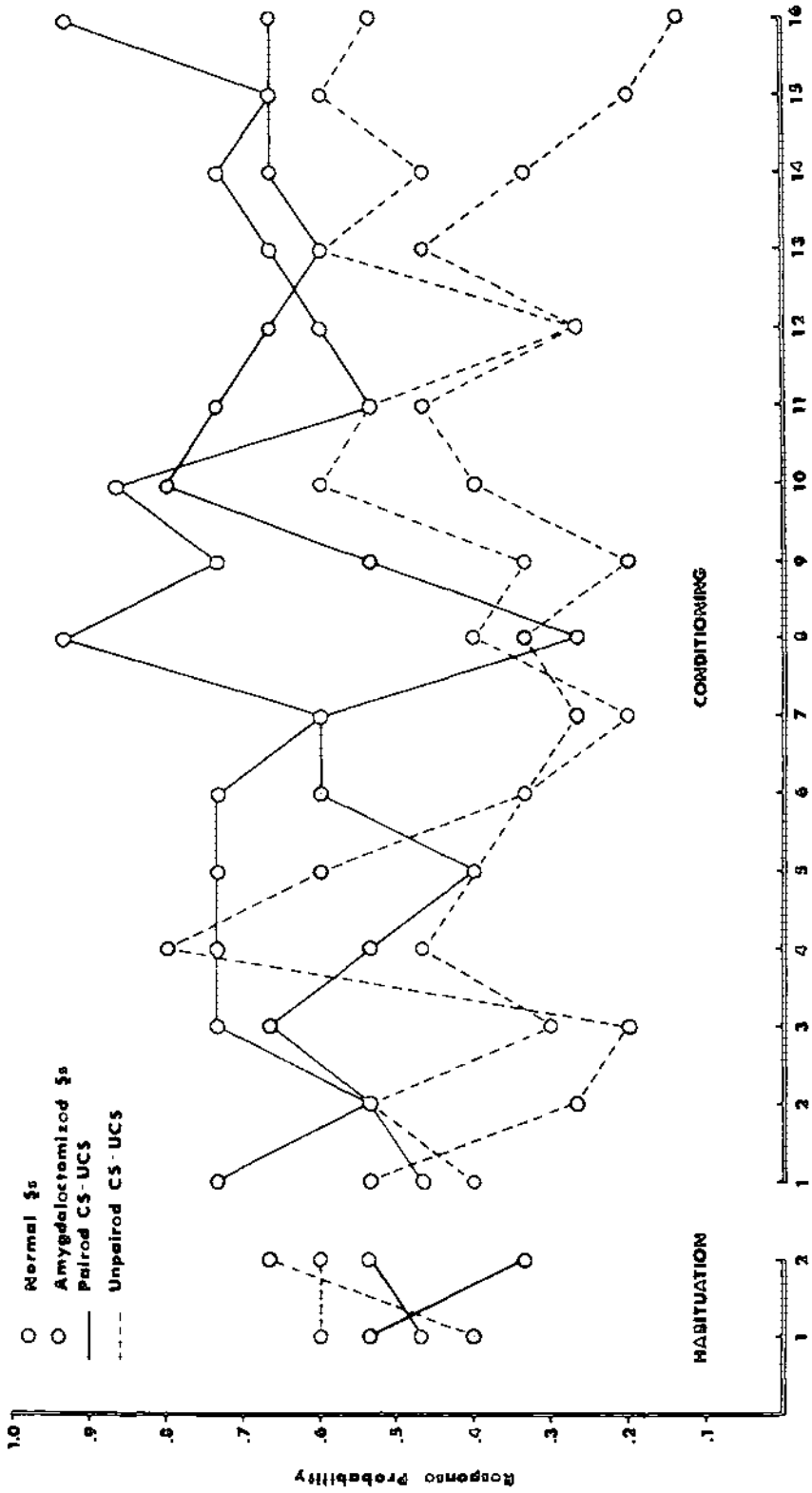
Normal Ss
Amygdalectomized Ss
Paired CS-UCS
Unpaired CS-UCS
HABITUATION
CONDITIONING
Response Probability
5 Trial Blocks

FIG. 6A

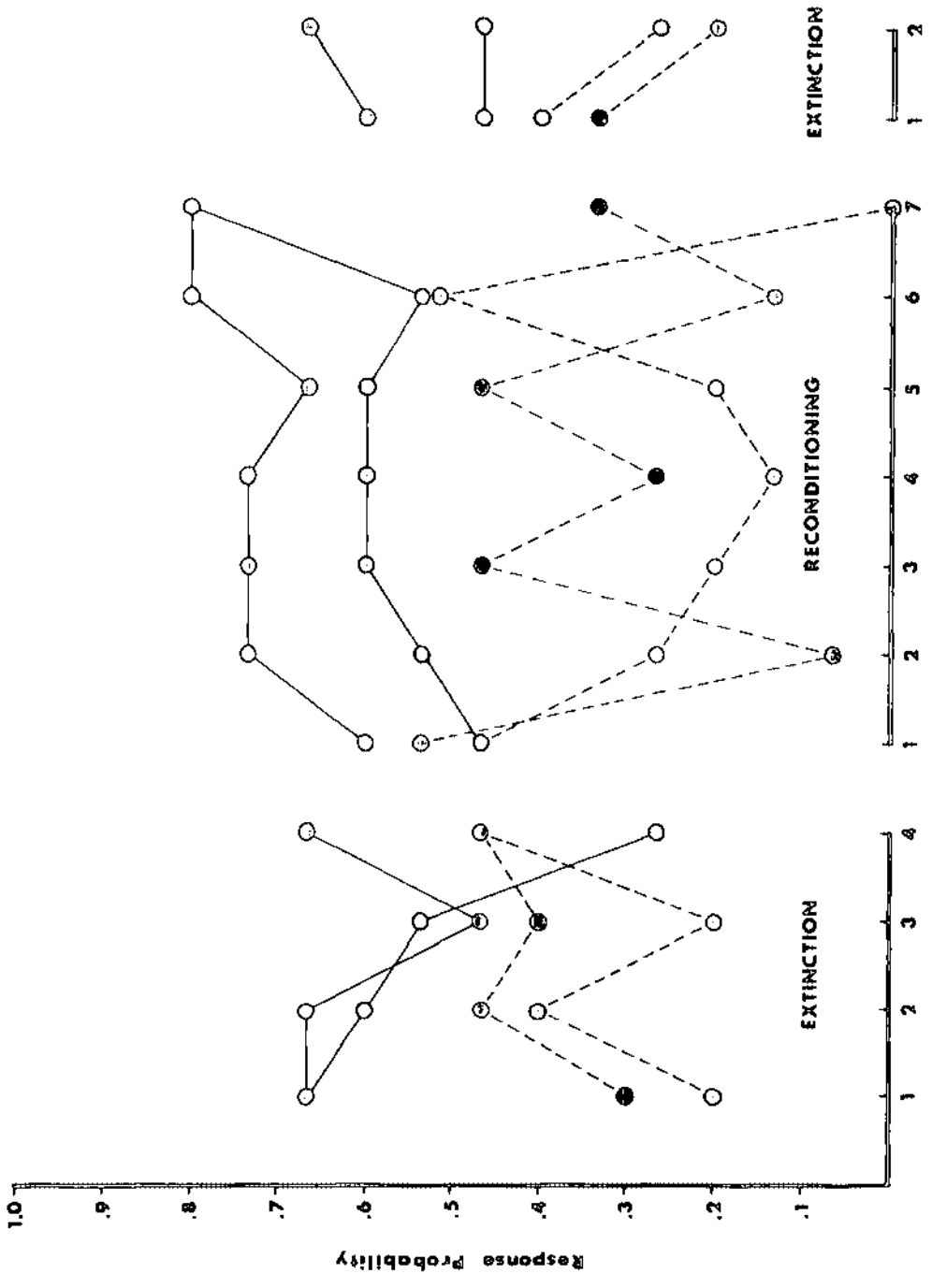
Response Probability
1.0
.9
.8
.7
.6
.5
.4
.3
.2
.1
EXTINCTION
1 2 3 4
RECONDITIONING
1 2 3 4 5 6 7
5 Trial Blocks
EXTINCTION
1 2

FIG. 6B

upon CS presentation. Moreover, these differences occurred only during the conditioning phases and were not apparent during the extinction phases when shock was no longer delivered during habituation. In addition, the nonsignificant lesions-by-conditions interaction effect in both frequency and amplitude analyses indicated that the amygdalectomized subjects, both paired and unpaired groups, exhibited responses that were as frequent and as great in amplitude as those of the corresponding groups with no lesions. Therefore, the procedures used in this experiment produced clear evidence of classical conditioning as opposed to pseudoconditioning or sensitization in both amygdalectomized and intact monkeys.

In this experiment, amygdalectomy had no significant effect on the *acquisition* of classically conditioned EMG or movement repsonses. No statistically reliable differences were found between the subjects with lesions and the intact subjects either in the amplitude or frequency of occurrence of conditional responses during any of the five phases of the experiment. Note that these are the same amygdalectomized subjects who *failed* to give evidence of conditional viscero-autonomic responses in Experiment 1.

Summary and Conclusion

The basic findings of these two studies are as follows: (1) a failure of orientating and conditioning of viscero-autonomic responses, but (2) essentially normal orienting and conditioning of a temporal muscle response in amygdalectomized animals. Small procedural differences exist between the two studies, and a possible order effect exists as a result of the use of the same subjects consecutively. However, we feel that the difference between the responses of the viscero-autonomic system and the skeletal system would still be found were all of the measures gathered simultaneously in an optimal conditioning situation. The definitive study—simultaneous viscero-autonomic and skeletal (behavioral) recording—remains to be done. Trial-by-trial analysis of the correlations or lack of correlations between these responses in such a study should be highly informative.

These two studies offer substantial information relevant to the original questions. When behavior is modified by simple repetition of experience the effect of amygdalectomy is restricted to the viscero-autonomic components of orienting and classical conditioning, not to the entire spectrum of responses. The issue raised, therefore, is the significance of the viscero-autonomic components of orienting and classical conditioning. The suggestion has been proposed (Pribram, 1969) that these components serve as mechanisms of internal rehearsal necessary to the registration (as novel or familiar) of the orienting and conditioning experience. This proposal requires further testing.

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