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Aerospace Medical Division
Air Force Systems Command
Holloman Air Force Base, New Mexico

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The animals used in this study were handled in accordance with the "Guide for Laboratory Animal Facilities and Care" as promulgated by the National Academy of Sciences - National Research Council.

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FOREWORD

This report is based on a study conducted jointly by the Florida State University, Tallahassee, Florida, and the 6571st Aeromedical Research Laboratory. The work was completed in August 1967 and was supported by United States Air Force contract number F29600-67-C-0012, project 6893, 6571st Aeromedical Research Laboratory, Holloman Air Force Base, New Mexico, and by contract number AT-(40-1)-2903 with the United States Atomic Energy Commission.

This technical report has been reviewed and is approved for publication.

C. H. Kratochvil

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Commander

ABSTRACT

By using a modified conditioned suppression technique, critical fusion frequencies were determined across 8.0 log units of brightness for three rhesus monkeys. Threshold values ranged from 20 c.p.s. for -3.9 log ft. L. to 95 c.p.s. for 4.1 log ft. L. Although systematic individual differences were observed at each brightness value, the intrasubject variability did not exceed 5 c.p.s. upon replication.

I

INTRODUCTION

Critical flicker frequency (cff) thresholds to photic stimulation in rhesus monkeys have previously been examined by investigators using either a two-lever tracking technique (1) or a simple discrimination design (2). When training monkeys using the tracking procedure, a flickering target serves as a discriminative stimulus (S^D) for lever A, and a continuously illuminated target serves as an S^D for lever B. With this procedure Symmes (1) determined cff thresholds across 4.0 log units of intensity, and the effect of light dark ratio was examined in five subjects. Variability of cff thresholds for the individual subjects was 10 c. p. s., and 20 to 40 percent of the data were rejected when controls for random switching behavior were instituted.

Mishkin and Weinkrantz (2), using 12 rhesus monkeys as subjects, reinforced lever pressing in the presence of a flickering light and withheld reinforcement when lever pressing occurred in the presence of continuous illumination. The subjects were also reinforced for non-responding during continuous illumination periods. The flickered and continuous illumination periods were randomly presented and the frequency of the flickered light was increased 5 c. p. s. per session when a high degree of discrimination had been observed. When the subject's lever pressing behavior during both the flickered and continuous illumination periods fell to chance level, the cff threshold was assumed to have been attained. However, it was found with the discrimination procedure that the cff threshold for individual monkeys systematically decreased. A change in the obtained threshold values as great as 25 c. p. s. occurred for several subjects over a year's practice period, and the cff was still increasing. This change in threshold value was said to be due to the influence of learning.

Hendricks (3) recently developed a technique for investigating cff thresholds in animals using a modified conditioned suppression design. She determined cff thresholds in pigeons as a function of stimulus intensity and obtained quite reliable thresholds. The threshold values seldom varied more than 3 c.p.s. from day to day over a period of several months. Powell (4), using the same technique and experimental subject, studied cff thresholds as a function of both intensity and pulse-to-cycle fraction. His data also indicated reliable threshold values.

The present study was designed to evaluate the conditioned suppression technique for determining the cff threshold in the rhesus monkey. Thresholds were studied as a function of the stimulus intensity over a range of 8.0 log units.

II

METHOD

A. SUBJECTS

Three male rhesus monkeys (Macaca mulatta) that had been previously used as control subjects in a match-to-sample study served as subjects. At the start of the experiment the ages of the subjects were estimated to be between 36 and 48 months.

B. APPARATUS

A Foringer primate chair was housed in a flat black 61 by 127 by 94 cm. (24 by 50 by 37 inches) plywood, sound-insulated chamber. White noise presented by means of a speaker in this chamber served as a masking sound to the subject. The stimulus light was displayed on a 4.4 by 5.1 cm. (1.75 by 2.0 inches) ground glass plate which was located in the horizontal plane approximately 19.1 cm. (7.5 inches) from the monkey's face. The stimulus light was the only source of illumination in the chamber with the exception of a small pilot lamp which served as a discriminative stimulus for the reinforcer, D&G[1] .7 g. whole diet monkey pellets. All reinforcement scheduling and stimulus presentations were carried out through a system of switching circuits and electric timers.

The optical system, which is shown in Figure 1, has been previously described in detail (4). The stimulus source (S) was a 620-watt iodine lamp operated on a high filtration d.c. power supply to minimize a.c. ripple. A 250 ml. flask (L_1) filled with distilled water served as a heat absorber and as a collimating lens. A photocell (P_1) connected

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[1] Dietrich and Gambrill, Inc., Foringer and Company, Inc., 535-A Southlawn Lane, Rockville, Maryland 20850.

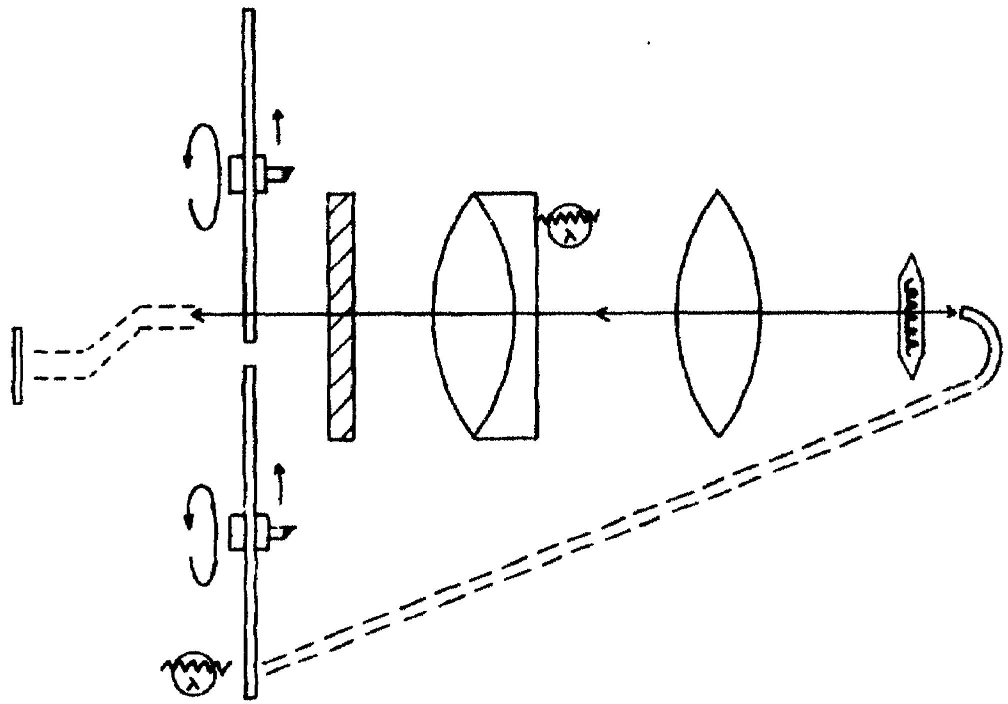

Figure 1. Diagram of the optical system: S, iodine lamp used as the stimulus source; L_1, 250 ml flask of distilled water which served as a heat absorbent and collimator lens; P_1, photovoltaic cell which allowed constant monitoring of the stimulus source intensity with a microammeter; L_2, imaging lens; D, neutral density filter; O_1, end of a fiber optic light tube providing an exciter light to monitor flicker frequency; F_1, constant speed rotating sectored disc; F_2, variable speed rotating sectored disc which provided the flickering stimulus when moved into the light path; P_2, photovoltaic cell which allowed continuous monitoring of the flicker frequency; 0_2, fiber optic light tube; G, plate of ground glass in the monkey chamber.

to a d.c. microammeter was located 1.9 cm. (.75 inch) from the water-filled flask, which permitted constant monitoring of the stimulus source. A constant light intensity was maintained by adjusting the voltage to the iodine lamp. The collimated light from the flask was focused by an imaging lens (L_2) on the end of a 91.4 cm. (3 foot) length of 0.64 cm. (0.25 inch) fiber-optics (O_2). The other end of the light pipe was placed 3.2 cm. (1.25 inches) from the back surface of the ground glass plate (G) in the monkey's chamber. This provided a stimulus spot approximately 2.8 cm. (1.1 inches) in diameter on the ground glass. With no neutral tint filters in the system, a brightness value of 4.1 log foot-Lamberts (log ft. L.) was measured on the ground glass by an SEI exposure photometer. Light intensity was varied by means of neutral density filters (D), and the flicker frequency of the stimulus was controlled by means of two motor-driven sectored discs (F_1 and F_2) located between the light tube and the imaging lens. A pulse-to-cycle fraction of .50 was used throughout all measurements. One of the sectored discs (F_1) was rotated at a constant speed, giving a frequency of 180 c.p.s. This disc was in the optical path at all times except when the flicker stimulus was presented. The second disc (F_2) was attached to a variable speed motor. By using a one-hole, three-hole, or six-hole disc, a frequency range of 1 to 150 c.p.s. was obtained. The change from the constant high frequency to the conditioning flicker frequency was accomplished by activation of a solenoid. A second fiber-optic light tube (O_1), transmitting light from the iodine lamp to one side of the variable frequency rotating sectored disc, was used to activate a second photocell (P_2) on the opposite side of this disc. The output of the photocell was connected to a frequency meter which was used to monitor all conditioning-stimulus frequencies. The output of the photocell was also connected to the x input of an oscilloscope, and the output of a Hewlett-Packard model 201c audio oscillator was fed into the y input of the oscilloscope. This provided a confirming standard for frequency monitoring via the lissajous pattern.

A Grason-Stadler model E6070B shock generator was used to provide the aversive stimulus which was paired with the conditioning stimulus. The shock was presented to the monkey by a foot electrode (5). All responses, reinforcements, and stimulus events were recorded on electronic counters, a cumulative recorder, and a four-pen event recorder.

C. PROCEDURE

The subjects were reduced to approximately 90 percent of their free feeding body weight. Each subject was shaped by the method of successive approximations to press a lever. Lever pressing behavior was initially maintained by continuous reinforcement and then by several values of the variable interval (VI) schedule of reinforcement. Each subject was gradually progressed from a VI 30-second to a VI 2 schedule. When stable response rates were established after approximately 25 sessions on the VI 2 schedule, conditioned suppression training was initiated.

During the initial suppression training, the stimulus source was flickered at a frequency of 20 c.p.s. for 30 seconds, and the flicker stimulus was terminated by an unavoidable shock. The intensity of the stimulus was 2.1 log ft. L. Responses during the 30 seconds prior to onset of the flicker stimulus and responses during the flicker stimulus were recorded. Quantification of suppression behavior was accomplished by the ratio suggested by Hoffman, Fleshler, and Jensen (6):

$$\frac{\text{(Pre-flicker Responses)} - \text{(Flicker Responses)}}{\text{Pre-flicker Responses}}$$

Using this suppression ratio, complete suppression is indicated by a value of 1.0 and no suppression by a value of 0.0.

Ten suppression training trials were presented daily to each subject. In order to ascertain that the monkey was suppressing to flicker rather than to some transient associated with the shift from one disc to the other, 10 control trials were also randomly presented during the training sessions. The control trials consisted of operating the solenoid and switching from the constant speed sectored disc to the variable speed disc for 30 seconds with the latter maintained at maximum rotation speed. Suppression training was continued for each subject until a criterion of 10 consecutive suppression ratios of .90 or above was reached.

When the suppression training criterion was achieved for a subject, cff threshold testing at the 2.1 log ft. L. stimulus intensity was initiated. The frequency of the flicker stimulus was increased 5 c.p.s. and the subject was tested for suppression at the new frequency. In several later threshold determinations, 2 c.p.s. increments in frequency were used instead of 5 c.p.s. During cff threshold testing, the criterion for suppression at each frequency was suppression ratios equal to or greater than .50 for three out of four trials. The flicker frequency of the stimulus was increased in discrete steps of 5 c.p.s. or 2 c.p.s. until cff threshold for the initial stimulus intensity was obtained. The cff threshold was defined as that frequency of the conditioning stimulus which failed to elicit suppression ratios of .50 or above on three out of four stimulus presentations. When cff threshold was reached, the procedure was repeated in order for the next stimulus intensity to be tested. A 60 percent partial shock schedule was used near threshold values in order to reduce the probability of shocking a subject below threshold and thus disrupting the stability of the baseline response rate. Subject was never shocked at frequencies near threshold when no suppression behavior was obvious. Each session 25 to 30 suppression trials were run and cff thresholds were obtained over a range of 8.0 log units of intensity. At least 13 different intensity values were used with each subject.

III

RESULTS

Polygraph recordings illustrating the typical procedure that was used during cff threshold determination are shown in Figure 2. The suppression behavior of M-392 at a stimulus intensity of 3.1 log ft. L. is illustrated by the tracings. Lever pressing behavior is almost completely suppressed during flicker presentation for frequencies of 62 c.p.s. and below. The amount of suppression is substantial even when the frequency of the flicker stimulus is only a few c.p.s. above threshold -- that is, at 64 and 66 c.p.s. No response suppression is evident, however, when the frequency is increased to 68 c.p.s. The suppression occurring during the control trial is negligible which indicates that suppression is controlled by the flicker stimulus and not by transients.

The precise nature of stimulus control, which is obtained during threshold determination by the conditioned suppression technique, is shown in Figures 3, 4, and 5. These graphs present mean suppression ratios as a function of frequency for subjects M-285, M-392, and M-299 for 14, 13, and 14 intensities respectively. High suppression ratios, typically .80 or above, were maintained for the lower frequencies, and an abrupt decrease in the suppression ratio is obvious as the frequency is increased. This characteristic pattern was observed for all subjects at the various intensity values investigated. The maximum ranges of mean suppression ratios for the control trials across all sessions for M-392, M-285, and M-299 were +.05 to -.06, +.05 to -.04, and +.11 to -.03 respectively.

In Figure 6 critical fusion frequency thresholds for each subject are plotted as a function of the intensity of the stimulus light. As can be seen in the figure, intersubject variability at high intensity levels was found. At stimulus intensities above -.90 log ft. L., however, essentially a linear function between stimulus intensity and fusion frequency was obtained for each subject.

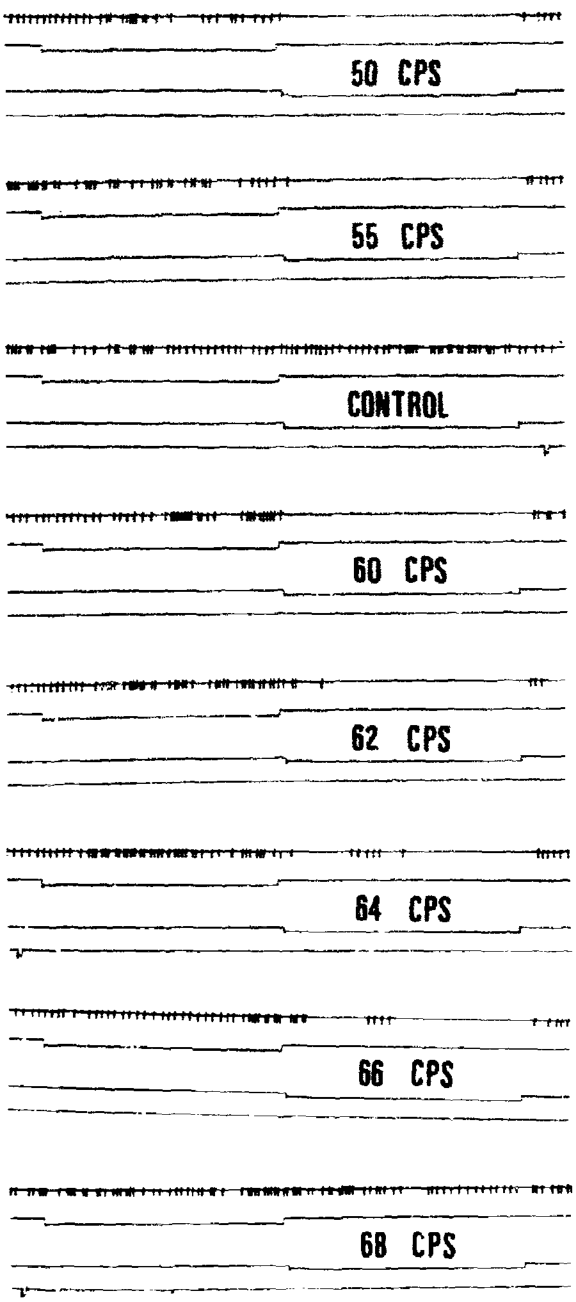

Figure 2. Polygraph recordings for subject M392 during cfi threshold determination with a stimulus intensity of 3.1 log ft. L. Responses are recorded on line 1; a 30-sec. pre-stimulus period on line 2; a 30-sec. flicker stimulus period on line 3; and reinforcements on line 4. Shocks were delivered with the termination of all conditioning stimulus frequencies except 68 c.p.s. Suppression ratios ranged from .97 to .91 for frequencies from 50 c.p.s. to 62 c.p.s. Other ratios were .84, .76, .13 and -.33 for 64, 66, 68 c.p.s. and control trial respectively.

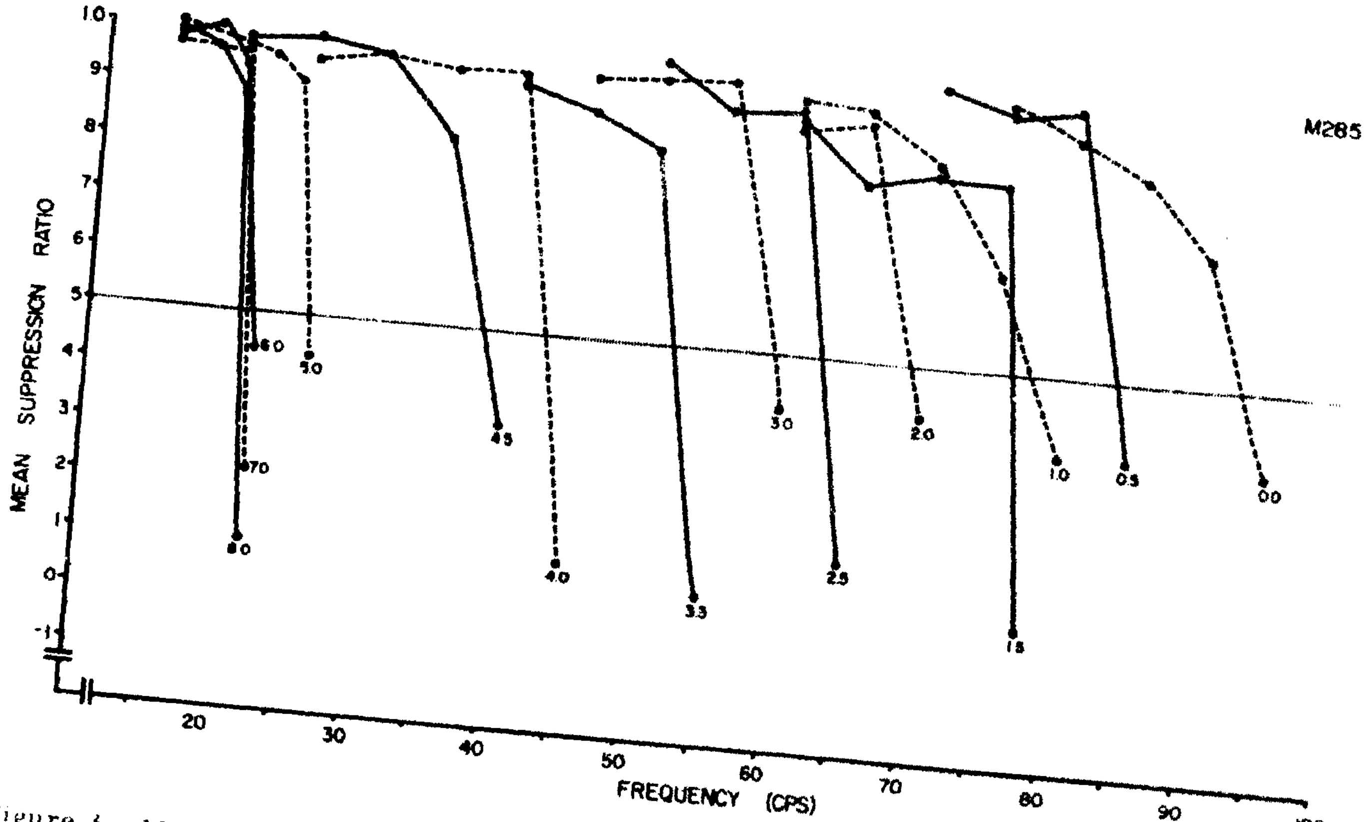

Figure 3. Mean of four suppression ratios as a function of the flicker frequency for subject M285 at 14 stimulus intensities. The stimulus intensity indicated by the curve labeled 0.0 equals 4.1 log ft. L. The off threshold at each intensity is the abscissa of the point below the .50 line.

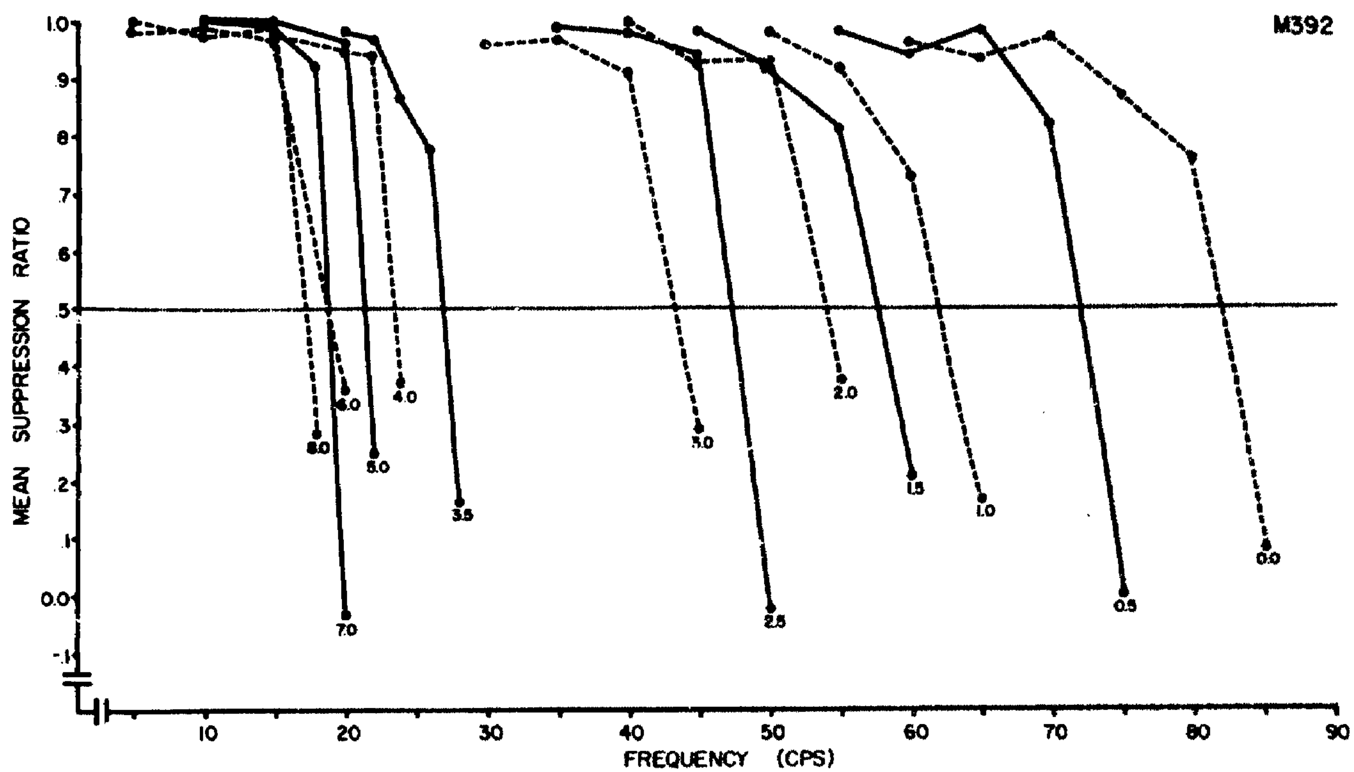

Figure 4. Mean of four suppression ratios as a function of the flicker frequency for subject M392 at thirteen stimulus intensities. The stimulus intensity indicated by the curve labeled 0.0 equals 4.1 log ft. L. The cff threshold at each intensity is the abscissa of the point below the .50 line.

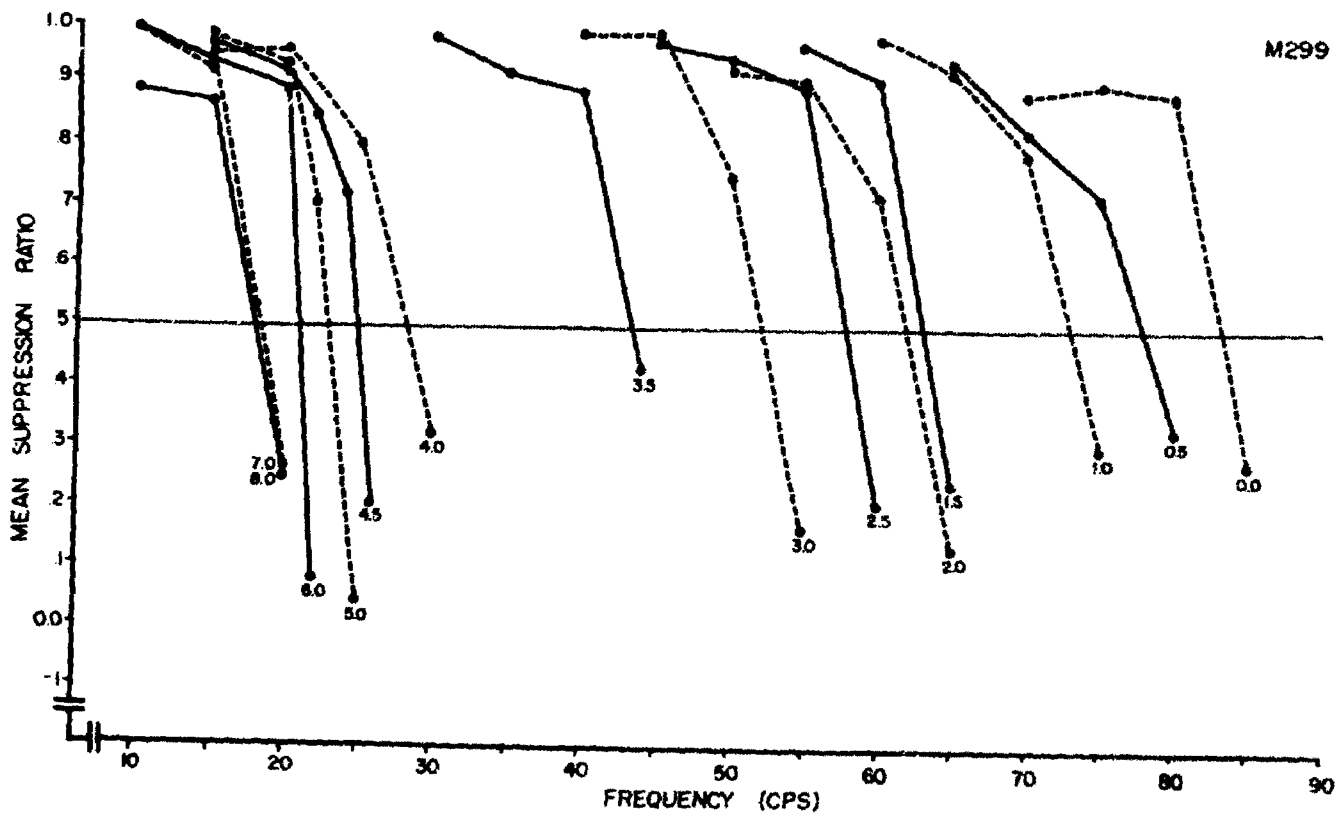

Figure 5. Mean of four suppression ratios as a function of the flicker frequency for subject M299 at 14 stimulus intensities. The stimulus intensity indicated by the curve labeled 0.0 equals 4.1 log ft. L. The cff threshold at each intensity is the abscissa of the point below the .50 line.

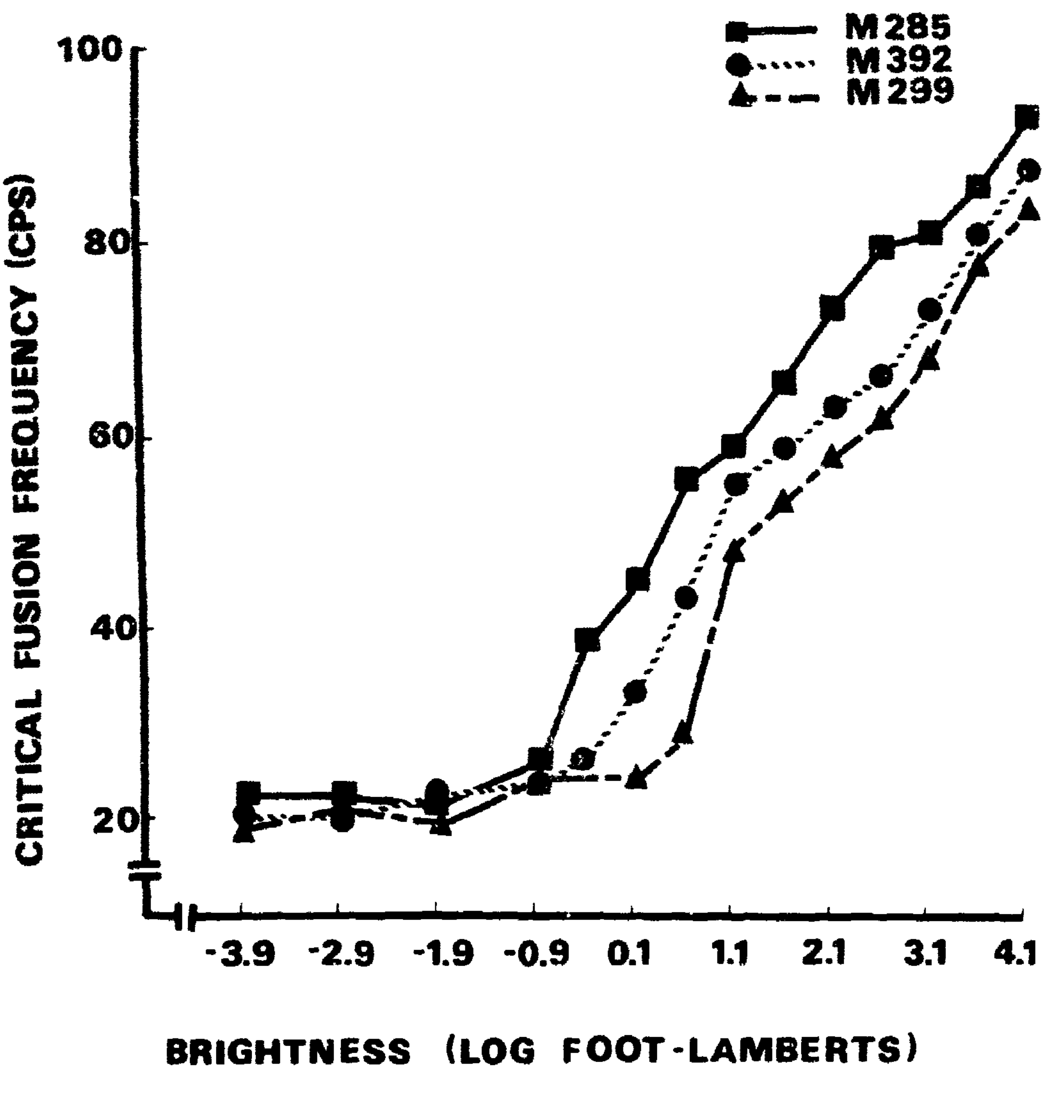

Figure 6. Critical fusion frequency thresholds for three subjects as a function of the intensity of the stimulus light.

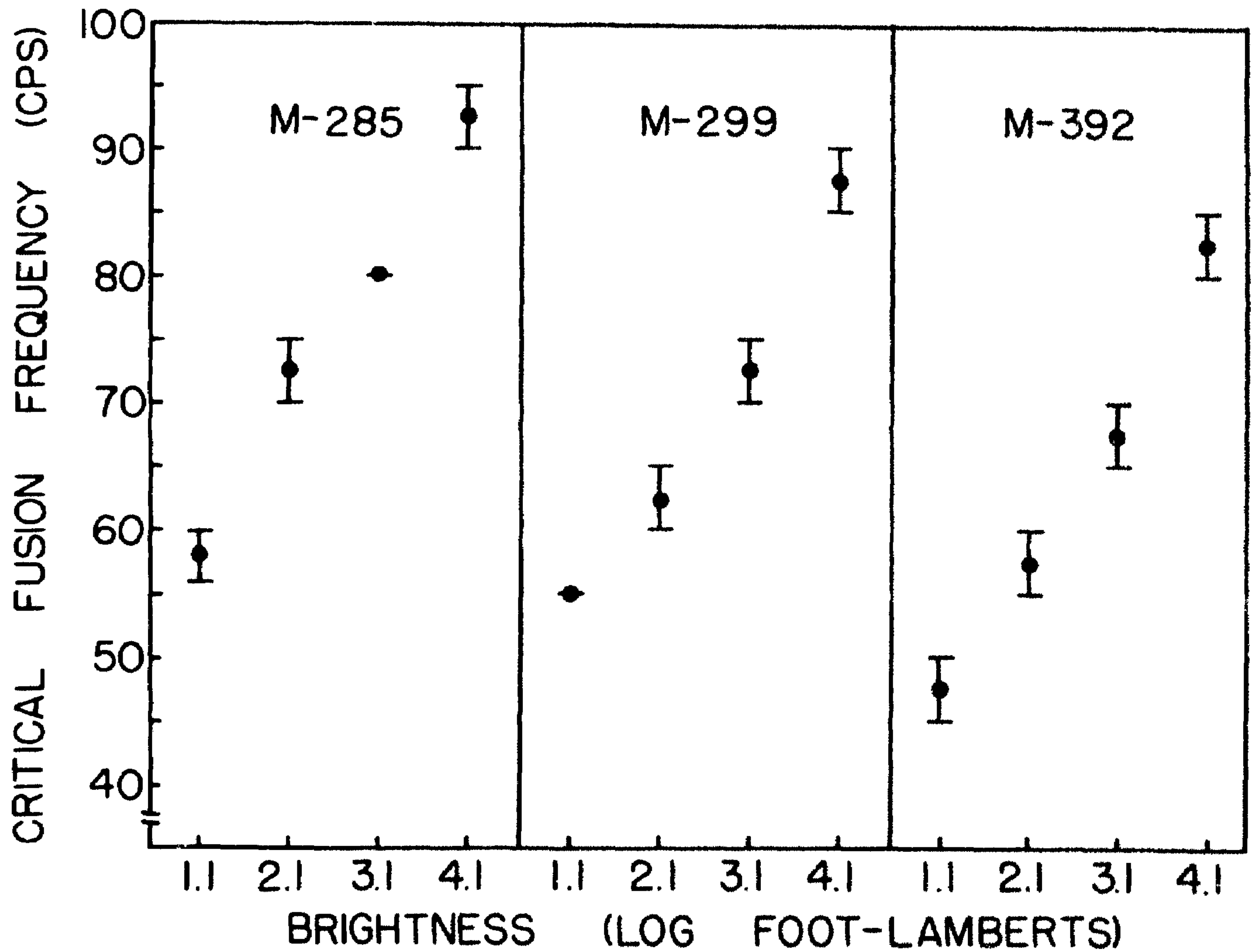

Figure 7. Critical fusion frequencies of the three monkeys for four intensities of the conditioning stimulus. Vertical lines drawn through the points represent the amount of observed variability for two threshold determinations. Where no vertical lines are drawn through *the points, the same threshold values were obtained upon replication.*

The degree of intrasubject variability for each subject at four intensity levels is shown in Figure 7. Each point represents the mean of two cff threshold frequencies determined on different days, and the vertical lines indicate the range between these two days. Intrasubject variability ranges from 0 to 5 c.p.s. upon replication.

IV

DISCUSSION

Methodological considerations of the conditioned suppression technique as a threshold procedure were discussed in detail by Hendricks (3) Factors considered were the efficiency of the technique in establishing and maintaining differential responding in the presence of the stimulus, the maintenance of stimulus control during threshold testing, the rigor of data analysis, intrasubject threshold reliability, and intersubject replication. Hendricks' data were collected using the pigeon as an experimental subject. The present study has extended the generality of the conditioned suppression technique as a threshold procedure to the rhesus monkey.

When a simple discrimination procedure is used to investigate thresholds in primates, responding during $S\Delta$ (periods when the S^D is not present) becomes a problem and loss of stimulus control is especially critical near threshold values. The low degree of intrasubject variability observed in the present study, however, exemplifies the degree of stimulus control obtained when the conditioned suppression technique is used with primates. Suppression of responding in the presence of a suprathreshold flicker stimulus is essentially complete, and the suppression behavior is generalized across the higher frequencies until threshold values are reached. Presenting a conditioning stimulus a few cycles per second higher than the cff threshold produces minimal or no suppression.

The efficiency of using aversive control superimposed on a stable baseline response rate as an animal psychophysical technique merits special attention. When a tracking procedure is used to determine thresholds in animals, switching behavior and adventitious reinforcement resulting from the use of two or more manipulanda become a major problem. Symmes (1, 7) found that when controls for switching behavior were introduced, a significant portion of his data had to be rejected.

With the conditioned suppression technique, a single lever is used and problems associated with dual manipulanda are avoided. In addition, retraining of a subject once suppression behavior has been established was unnecessary during the present study, but other investigators have reported the necessity of retraining (1).

The results of the present investigation indicate that the conditioned suppression technique is more reliable and more efficient for determining cff thresholds in rhesus monkeys than either the tracking procedure or the simple discrimination design. Symmes (1) reported that intrasubject thresholds obtained using the tracking design varied by 10 c.p.s., and Mishkin and Weinkrantz (2) found that cff thresholds varied as much as 25 c.p.s. over a period of one year. The cff threshold obtained in the present study with the conditioned suppression technique did not vary more than 5 c.p.s. for any animal over a continual 6-month testing period.

All animals in the present study showed a linear relationship between cff thresholds and the intensity of the stimulus which is in accord with the Ferry-Porter law, the primate data of Symmes (1) and the pigeon data of Hendricks (3). No subject showed an appreciable change in cff value below the stimulus intensity of -.90 log ft. L. Presumably, this flattening of the cff threshold for the lower intensities is due mainly to a contribution of the rod receptors.

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13. ABSTRACT

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