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Effects of d-Amphetamine and Methylphenidate upon Auditory Threshold in the Squirrel Monkey[1]

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DELAY, E. R., N. O STEINER AND W. ISAAC. *Effects of d-amphetamine and methylphenidate upon auditory threshold in the squirrel monkey.* PHARMAC. BIOCHEM. BEHAV 10(6): 861–864, 1979.—The effects of d-amphetamine sulfate and methylphenidate hydrochloride on auditory thresholds in ten squirrel monkeys were examined using a 4.2 kHz stimulus in a free field. The results indicated that d-amphetamine raised auditory thresholds but methylphenidate did not alter the thresholds. The elevation of sensory thresholds by d-amphetamine was in agreement with previous studies suggesting that the drug acts as a behavioral depressant in diurnal animals.

Squirrel monkey Auditory threshold d-Amphetamine sulfate Methylphenidate hydrochloride

SENSORY input [17] and pharmacological agents [3], such as d-amphetamine sulfate (Dexedrine), have been shown to influence reticular arousal. Isaac and Devito [10] hypothesized that the behavioral reflection of arousal could be modified by varying the intensity of ambient sensory input to the reticular formation. Diurnal squirrel and rhesus monkeys, shown to be more active in the light than in the dark [1, 10, 11, 15], responded to d-amphetamine with dose related decreases in activity when tested in the light but exhibited no change in activity levels when tested in the dark [11,15]. Alexander and Isaac [1] have suggested that d-amphetamine alters the arousal level of an organism by reducing the arousal effects of elevated ambient illumination. Therefore the drug would have an apparent depressant effect in diurnal animals.

Factors capable of modifying arousal have also been shown to influence sensory thresholds [4, 7, 12, 18]. It would appear that increased ambient sensory stimulation lowers sensory thresholds whereas d-amphetamine seems to elevate thresholds in diurnal animals.

A drug which appears to affect behavior in a manner similar to d-amphetamine is methylphenidate hydrochloride (Ritalin). Clinically, methylphenidate and d-amphetamine are used interchangeably for treatment of hyperkinesis in children [16]. Kallman and Isaac [13] compared the effects of d-amphetamine with that of methylphenidate on locomotor activity of rats tested in the light and the dark. The effects of both drugs interacted with the level of ambient illumination, each drug producing greater increases in activity in the light than in the dark, but methylphenidate required approximately twice the dose to produce an equivalent behavioral effect. Davis [5] reported that equivalent doses of methylphenidate and racemic amphetamine reduced pacing behavior in rhesus monkeys in a similar manner.

The present study investigated the effects that d-amphetamine and methylphenidate may have upon auditory thresholds. If d-amphetamine reduces the effectiveness of illumination upon arousal in the squirrel monkey, then auditory thresholds should increase in a dose-related manner. Similar predictions would be made for methylphenidate if it is effecting the organism in a fashion similar to d-amphetamine.

METHOD

Animals

The animals were five male and five female naive squirrel monkeys (*Saimiri sciureus*). The animals were two years old at the beginning of the experiment and had been in the laboratory for one year. The monkeys were fed a normal colony diet consisting of moistened Purina Monkey Chow, fresh lettuce, fruit and a vitamin supplement. To maintain a constant, low drive level, the daily food ration was divided and presented in two feedings with water available ad lib in the home cage. The animals were fed 4 hr before testing, tested between 9:00 a.m. and 2:00 p.m., and fed again about 2 hr after testing. The average weight of each animal was 572 g at the beginning and 604 g at the end of the experiment, showing a normal growth rate compared to other squirrel monkeys of this age in the colony. Colony illumination was maintained on a 12 hr light-12 hr dark cycle with the lights turned on at 5:00 a.m.

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Apparatus

The animals were transported to the experimental apparatus and tested in an expanded metal carrying cage 25.4×25.4×40.6 cm. The doors at both ends of the transport cage were made of Plexiglas painted black. A lever (Davis, Model BD-2) was inserted through a small opening in one of the doors. The lever served as both the response manipulandum and as the reinforcement site.

The animals were placed in a sound attenuated environmental test chamber (BRS/LVE No. 133-10). A frame of acoustical tile and fiberglass insulation was constructed around the chamber to improve its sound attenuating properties. An incandescent light provided 25 ft-c. illumination in the center of the carrying cage. Masking noise from a ventilation fan was sampled in the center of the free field with a General Radio Co. Graphic Level Recorder (Type 1521-B) and analyzed with a General Radio Co. Wave Analyzer (Type 1900-A). The analysis of the masking noise showed that the intensity of the masking noise was 50 db (re: 0.0002 dynes/cm^2) at 100 Hz and decreased to 20 db at 1000 Hz. Between 1000 and 8000 Hz the intensity dropped gradually to slightly below 15 db. The sound pressure of the masking noise at the stimulus frequency was 15 db. The stimulus was a 4200 Hz tone produced by integrated circuitry [6]. The sound pressure levels of the stimulus were measured at the center of the free field using a Bruel and Kjaer Sound Pressure Meter (Type 2203) coupled with a Bruel and Kjaer Octave Filter Set (Type 1613). Eight intensities were used: 14, 15, 17, 18, 18.9, 20, 21 and 25 db above the noise level at that frequency. The stimulus was presented through a 4 Ohm speaker for a duration of 500 msec on each trial. Each intensity was presented in random order three times under the experimental condition of each daily session. All programming and recording equipment was located in a separate room.

Procedure

The animals were initially trained to press the lever for sweetened Hawaiian Punch. Subsequently, the reinforcer was available only during the presentation of the auditory stimulus. To facilitate acquisition of this discrimination, a cue light was initially paired with the auditory signal, and then was gradually phased out after performance stabilized. When the animals showed consistent response latencies of less than 3 sec to the auditory signal, the intensity of the stimulus was decreased until the animals failed to respond. The threshold value, defined as the sound pressure level corresponding to the 50% response level, was estimated. Eight stimulus intensities surrounding the threshold value were selected to include performance levels ranging from over 90% to less than 10% stimulus detection. After the stimulus values had been selected, all of the animals were then trained for an additional month to ensure stable detection performance and fewer than 100 false alarms per session.

Bacteriostatic water was used as placebo and as the vehicle of the drug doses, all of which were prepared in constant volume solutions. Placebo, d-amphetamine sulfate (0.1, 0.2 or 0.4 mg/kg) or methylphenidate hydrochloride (0.4, 0.8 or 1.6 mg/kg) was administered orally in 9 ml of sweetened Hawaiian Punch 15 min prior to transporting the monkey to the chamber each day. The animal was adapted to the test conditions in the chamber for 5 min. During this period four warmup trials, not included in the analysis, were presented using the highest stimulus intensity. A trial began with the stimulus onset and was 3 sec in duration. A correct response, or detection, was a lever press made at any point during the trial. A detection terminated the trial and delivered 1/8 ml of sweetened Hawaiian Punch. Incorrect responses, or false alarms, were recorded cumulatively for a complete experimental session with the exception of a 10-sec period following each delivery of the reinforcer. This delay eliminated recording lever presses resulting from the animal obtaining the reinforcer. The intertrial intervals were randomized and ranged from 30 sec to 110 sec, with a mean of 70 sec. To facilitate stimulus control and ensure a low level of false alarm responding, the onset of the stimulus was delayed for 10 sec if the animal emitted a lever press during the 10 sec prior to the stimulus onset. In addition, if an animal had over 100 false alarms during a session, the data for the day were disregarded and the animal was retested under the same drug dose at the end of the replication. This procedure, however, was necessary on only 3 occasions.

Half of the animals, 2 males and 3 females, began the experiment with d-amphetamine and the other half of the animals, 3 males and 2 females, began with methylphenidate. The order of doses for each drug was counterbalanced with a different order used in each replication. The first 3 replications were used to adapt the animals to the taste of the drug in the punch solution, so that they would reliably accept the drug, and to any changes in internal stimulus conditions of the animals related to the nonspecific effects of the drugs. Data from 4 subsequent replications were used in the analysis. After completing the last replication of the first drug, the animals were run for 6 days in the same procedure, but without drug treatment. At the end of this period the animals began a new sequence of 7 replications with the second drug. Again, the first 3 replications were for drug adaptation and the last 4 were for analysis. In this way all the animals were tested under both drug conditions in identical fashion in a counterbalanced order.

Any changes in performance with increasing doses of d-amphetamine could be the result of the anorexogenic properties of the drug reducing drive and, consequently, the amount of the reinforcer that the animal would consume. To examine this possibility, upon completion of the threshold study each of 3 pretest conditions was administered one time to each animal in random order over a 3 day period. The animals were given placebo in condition one and 0.4 mg/kg in condition two with both doses administered in a 9 ml of sweetened Hawaiian Punch. For the third pretest condition, the animals were allowed to consume sweetened Hawaiian Punch to satiation. Each solution was administered to the animals 15 min prior to being transported to the chamber for threshold testing. At the end of the test session, sweetened Hawaiian Punch was made freely available to each animal and the amount of punch consumed at this time was recorded for analysis.

RESULTS

The total number of detections for the 4 replications following adaptation were summed and these totals were analyzed. Analysis of variance examining the effects of the 4 dose levels upon 8 intensity levels and sex of the animals was used in the evaluation of each drug.

Analysis of variance did not reveal any significant difference between detection rates under the placebo conditions of

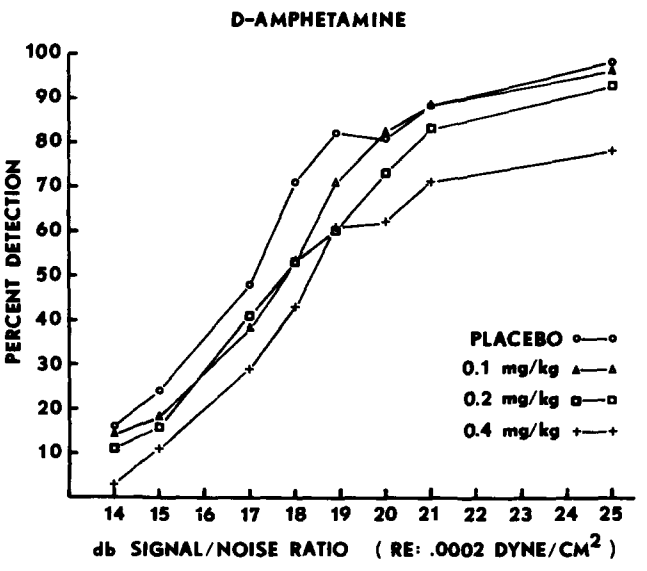

FIG. 1. The effects of d-amphetamine upon auditory thresholds at 4.2 kHz

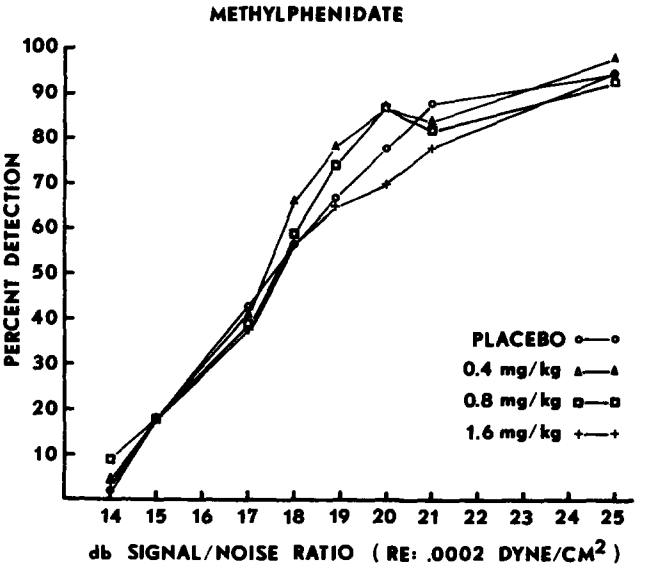

FIG. 2. The effects of methylphenidate upon auditory thresholds at 4.2 kHz.

the two drug regimens, $F(1,18)=2.63$, $p>0.10$. When the data from the 2 placebo conditions were combined, the auditory threshold was slightly greater than 17 db. This appears to be consistent with previous findings where squirrel monkey audibility functions at the frequency used in this study were measured in free field conditions [2], as well as with headphone equipment [9]. Differences in detection rate for the 8 stimulus intensities were significant for both d-amphetamine, $F(7,56)=151.11$, $p<0.001$, and methylphenidate, $F(7,56)=173.46$, $p<0.001$.

Significant dose related changes in thresholds, $F(3,24)=8.69$, $p<0.001$, were observed with d-amphetamine. Analysis of the mean number of detections made at each of the 4 dose levels of d-amphetamine with the Duncan Multiple Range test revealed that performance under the placebo condition was significantly different from 0.1 mg/kg ($p<0.05$) as well as 0.2 and 0.4 mg/kg ($p<0.001$). The difference between 0.1 and 0.2 mg/kg was not significant, but detection under 0.4 mg/kg of the drug was significantly different from all other doses ($p<0.001$). A dose by sex by intensity interaction was significant, $F(21,168)=2.45$, $p<0.01$, with males being more sensitive to the drug than the females. Methylphenidate did not have a significant effect on auditory thresholds, $F(3,24)=1.69$, $p>0.10$.

The mean number of false alarms during a session per monkey was 38.72 (SD=21.93). Analysis of variance using partitioned error terms did not reveal any systematic effect due to drug, dose or sex of the animal on false alarm responding.

Analysis of the data examining the influence of d-amphetamine on punch intake indicated that the animals consumed a mean of 16.3 ml of punch after administration of the placebo solution and 13.0 ml following administration of the drug solution. Mean punch consumption after the satiation condition was 1.2 ml. Punch consumption following the administration of the drug solution did not significantly differ from punch consumption following the placebo condition ($t=0.50$; df=9; $p>0.10$). However, punch consumption following satiation was significantly different when compared to the placebo condition ($t=2.27$; df=9; $p<0.05$).

DISCUSSION

The threshold data obtained under the placebo conditions for the 4200 Hz tone used in this study appear to be in agreement with audiograms reported for the squirrel monkey by Beecher [2] and Green [9]. The auditory thresholds were elevated with increasing doses of d-amphetamine. One possible explanation for this increase is that the drug may have been producing changes in motivation as a result of its anorexogenic properties. This seems unlikely since the administration of 0.4 mg/kg d-amphetamine did not significantly reduce the amount of reinforcer consumed by the animals at the end of the test session. It is also possible that a drug induced decrease in response rate may have produced changes in detection rate under d-amphetamine. Another alternative explanation is that the drug may have increased response latencies of the animals beyond the 3 sec limit of a trial. Since all responses not made during the 3-sec trials were counted as false alarms, with the exception of 10 sec following a detection, response latencies longer than 3 sec due to the drug would have produced an increase in false alarm rate. However, analysis of the false alarm data indicated that these responses were not significantly altered by d-amphetamine and suggests that neither a decrease in response rate nor increased response latencies can account for the shifts in detection rate under the drug.

Lindsley [14] postulated that variations in the arousal level of an organism would influence ongoing perceptual processes. It has been suggested that behavioral arousal can be manipulated by altering ambient sensory conditions [10].

Investigations examining the effect of ambient illumination on tactile thresholds of humans [12] and rhesus monkeys [4] have found the thresholds lower in the light than in the dark. Furthermore, with test conditions similar to those employed in this paper, Delay, Smith and Isaac [7] reported that auditory thresholds for squirrel monkeys were also lower in the light than in the dark. These studies suggest that ambient illumination has a facilitory effect on other sensory modalities in diurnal species.

In contrast, it has been hypothesized that d-amphetamine reduces the effect of ambient illumination upon arousal [1]. Administration of d-amphetamine to rats produced dose related increases in locomotor activity which were greater in the light than in the dark [13]. The interaction of d-amphetamine and illumination eliminated the light-dark differences in activity. On the other hand, when the drug was administered to diurnal monkeys, activity decreased in the presence of light [1,11]. Additional support for the hypothesis was found when Isaac and Troelstrup [11] compared the influence of d-amphetamine on activity in nocturnal owl monkeys and diurnal squirrel monkeys. They found that the drug increased the activity of the nocturnal monkey and decreased the activity of the diurnal monkey in the light. However, the drug did not have an effect on either species in the dark. The drug appears to have the same effect on behavior as reducing illumination and, therefore, acts as a stimulant in nocturnal species, and a depressant in diurnal species. Thus a more plausible explanation for the increased thresholds is that d-amphetamine may be reducing the effects of illumination upon the arousal level of the monkeys.

If d-amphetamine is a depressant in diurnal monkeys in the presence of light, then sensory thresholds would rise with increasing doses in a manner similar to threshold changes seen with a reduction of illumination level [4, 7, 12]. Previous threshold investigations have shown that d-amphetamine produced threshold increases in visual [18] and tactile [4] modalities in rhesus monkeys. The results of this study add further support to this position and extend it to the auditory system.

Methylphenidate did not appear to have an effect on auditory thresholds in this study. This was somewhat surprising in view of previous reports using doses of methylphenidate within the range used in this study. Goethe and Isaac [8], in a literature survey of clinical dose ranges for d-amphetamine and methylphenidate, found that doses as low as 0.1 mg/kg have been reported to produce behavioral changes in humans. Davis [5] has reported that subcutaneous injections of methylphenidate were capable of reducing pacing behavior in rhesus monkeys with doses of 0.5 mg/kg or greater. Using intraperitoneal injections, Kallman and Isaac [13] compared the dose response curves of d-amphetamine and methylphenidate on locomotor activity of rats tested in the light and dark. These investigators found that d-amphetamine appeared to be twice as potent as methylphenidate. The doses of methylphenidate used in this study were four times the concentration of the doses of d-amphetamine that altered auditory thresholds when administered orally. However, oral administration of methylphenidate did not alter threshold responding, even with doses of methylphenidate much higher than those known to alter behavior with other modes of administration. This would suggest that generalizations concerning the effect, or lack of effect, of methylphenidate may be limited by the route of administration of the drug.

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