Pergamon

Pharmacology Biochemistry and Behavior, Vol. 50, No. 1, pp. 115-119, 1995

0091-3057/95 $9.50 + .00

0091-3057(95)00304-1

Drug Discrimination Training With Low Doses: Maintenance of Discriminative Control

ARTHUR TOMIE,[1] PENNY L. SHULTZ, MICHAEL S. SPICER AND LAURA L. PEOPLES

Department of Psychology (Busch Campus), Rutgers University, New Brunswick, NJ 08903

Received 31 May 1994

TOMIE, A., P. L. SHULTZ, M. S. SPICER AND L. L. PEOPLES. *Drug discrimination training with low doses: Maintenance of discriminative control.* PHARMACOL BIOCHEM BEHAV **50**(1) 115-119, 1995.—Procedures are reported that maintain control by the drug cue during and after drug discrimination training with lower doses that yield predominantly vehicle-appropriate choices. Twelve pigeons were trained to discriminate chlordiazepoxide (CDP) from saline using two-key (drug vs. vehicle) drug discrimination procedures. Intermixed within each block of 30 sessions were nine sessions of training with 8.0 mg/kg CDP, nine with one of seven lower training doses (4.0, 2.8, 2.0, 1.4, 1.0, 0.7, or 0.5 mg/kg CDP), and 12 with saline. The lower training dose was decreased across blocks. The three lowest training doses (1.0, 0.7, and 0.5 mg/kg CDP) yielded predominantly saline-appropriate choices but had no effect on discrimination of 8.0 mg/kg CDP or saline. Three doses (2.0, 1.4, and 1.0 mg/kg CDP) were retrained, and each yielded percentages of drug-appropriate choices nearly identical to those obtained during previous training. This drug discrimination procedure maintains control by the drug cue during and after training with vehicle-like doses of the training drug and may allow for repeated assessment of effects of low training doses.

Drug discrimination Training procedures Threshold Sensitivity Chlordiazepoxide Pigeons

DRUG discrimination procedures have been widely used to characterize drugs on the basis of their stimulus properties. In this procedure, subjects are injected presession with either drug or vehicle, and then reinforcement during the session is made contingent on injection-appropriate choice behavior (e.g., left-key responding is reinforced after drug administration, whereas right-key responding is reinforced after vehicle administration). Investigators evaluating the effects of progressively decreasing the training dose within-subjects (i.e., fading technique) have reported that the percentage of drug-appropriate choices on drug sessions decreases as a function of training dose. This relationship has been reported in rats trained to discriminate cocaine (4), amphetamine (18,25), morphine (21,23), fentanyl (5), phencyclidine (3), quipazine (1), LSD (8), pentylenetetrazol (10), and scopolamine (13); in pigeons trained to discriminate morphine (17) and phencyclidine (14); and in humans trained to discriminate caffeine (9,22).

Researchers employing fading techniques have reported that training with doses that yield predominantly vehicle-appropriate choices reduces subsequent discriminative control by higher training doses (15,17). For example, Peoples reported that after discrimination training with subthreshold training doses of morphine, reliable discriminative control by higher morphine training doses could only be reinstated by extensive retraining with those higher doses (17). In the most extensive evaluation of retraining reliability, Overton reported that in five of seven subjects he was unable, despite extensive retraining, successfully to reestablish discriminative control by doses of the training drug that had been accurately discriminated before training with the low doses (15).

The loss of discriminative control by the drug cue may be due to extended training with doses of the training drug that yield predominantly vehicle-appropriate choices. During this training, subjects presumably experience vehicle-like cues on both drug and vehicle training sessions and, consequently, provide few drug-appropriate choices, even on drug training sessions. During training with vehicle-like doses, therefore, vehicle-appropriate choices are often followed by extinction, and virtually all of the reinforcers are delivered after vehicle-appropriate choices. For these reasons, extended training with vehicle-like doses may obscure previously learned contingencies relating reinforcement to interoceptive discriminative stimuli, resulting in the deterioration of subsequent discrimination performance.

The present study assesses maintenance of discriminative

[1] To whom requests for reprints should be addressed.

control by the drug cue during and after appraisals of vehicle-like doses of the training drug. Our drug discrimination procedures differed from those employed by previous investigators in that training sessions with a highly discriminable dose of the training drug are intermixed with training sessions with lower doses and with vehicle. Specifically, the procedures employed intermix training sessions providing for presession injections of either: a) a relatively high dose of the training drug (8.0 mg/kg chlordiazepoxide, CDP); b) a lower dose of the training drug (4.0, 2.8, 2.0, 1.4, 1.0, 0.7, or 0.5 mg/kg CDP); or c) saline. Within each 30-session block, the 8.0 mg/kg dose of CDP and the lower dose of CDP were each administered before nine sessions, whereas saline was administered before 12 sessions. Only one of the lower training doses was given during a block of 30 sessions, and this dose was progressively decreased across blocks. Thus, the order of introducing the lower training doses was 4.0, 2.8, 2.0, 1.4, 1.0, 0.7, and 0.5 mg/kg CDP, as indicated earlier.

METHOD

Subjects

Subjects were 12 adult homing pigeons obtained from a local supplier and housed individually in metal and Plexiglas cages in a colony room with a 12-h (on at 0800 h) light–dark cycle. Pigeons were maintained at 80% of their free-feeding body weights and had free access to grit and water in their home cages. The free-feeding weights of the birds ranged between 440 and 568 g, with a mean of 501 g.

Apparatus

Apparatus consisted of eight standard three-key operant conditioning chambers (35 × 30 × 30 cm) for pigeons, enclosed in sound-attenuating casings. The operant chambers consisted of three response keys (each 2.9 cm in diameter) mounted horizontally on the front metal panel 20 cm above the metal grid floor. The center key (not used during this study) was 19 cm above the food-hopper aperture (6.35 × 6.35 cm). The two side keys were 3.5 cm to the right and left of the center key and were equipped with Industrial Electronic Engineers in-line display cells containing GE 1815 miniature lamps. Illumination of these lamps projected a white vertical line on the otherwise dark response keys. Two pairs of houselights were mounted behind a 2.4-cm-wide piece of Plexiglas directly above the front panel. The inner light of each pair (equipped with GE 74 bulbs) provided ambient illumination throughout experimental sessions while the outside member of each pair was only illuminated during discrimination training trials. Ventilation and masking noise were provided by an exhaust fan positioned behind the front panel. Four operant chambers had response key assignments drug-left, saline-right, whereas the remaining four chambers had the opposite key assignments. Session events were determined and data were collected by standard relay equipment and Commodore PET microcomputers.

Procedure

Preliminary training. The 12 pigeons were randomly assigned to two groups. For one group of six pigeons, the right key was CDP-appropriate and the left key was saline-appropriate. These assignments were reversed for the remaining group of six pigeons. During all phases of the study, sessions were conducted 6–7 days/week. Pigeons were allowed to locate and eat grain from the food hopper, then trained to peck on the response keys for food. Drug discrimination training was initiated when subjects were responding on a fixed ratio 10 (FR10) schedule on both keys.

Preliminary drug discrimination training. To minimize the disruptive effects of CDP on motor performance and feeding, initially drug discrimination training was conducted using a 4.0-mg/kg training dose of CDP, and throughout the study injections were given 1 h before the start of the session. Subjects were trained to discriminate a presession intramuscular (IM) injection of a 4.0-mg/kg dose of CDP from a presession IM injection of a 1-ml/kg volume of 0.9% (saline) solution. All injections were given in the breast muscle, after which subjects were returned to their home cage. At the beginning of each session, both pairs of houselights were illuminated, signaling a discrimination training trial, and both the left and right response keys were illuminated until the subject made 10 responses on one of the keys. If the subject made a total of 10 responses on the injection-appropriate key before doing so on the alternative key, the choice was recorded as correct, regardless of the number of responses (zero to nine) completed on the injection-inappropriate key. Illumination of the alternative key was terminated, and the subject was presented with 3-s access to a hopper tray filled with mixed pigeon grain. Moreover, the subject was provided with nine more opportunities to procure food on an FR10 schedule. If, on the other hand, the subject first completed the FR10 requirement on the injection-inappropriate key, then illumination of the alternative key was terminated and food was not presented during a 5-min period of extinction. The extinction period was followed by a 30-s intertrial interval.

Regardless of the correctness of choice on the first trial of each session, a total of three choice trials were scheduled during each session. Thus, it was possible for each subject to procure a maximum of 30 food presentations per session. The procedures used during the second- and third-choice trials were identical to the first. During the 30-s interval between trials, the outer member of each pair of houselights was turned off and neither key was illuminated. The session was terminated when the subject completed three trials or when 15 min had elapsed from the beginning of the session. Only the data from the first choice trial of each session were used to evaluate discriminative control by the drug. The data recorded were first trial choice (drug-appropriate or saline-appropriate), which was defined as the response key on which the subject first completed ten responses. When the group mean percentage of drug-appropriate choices on drug sessions was at least 80% for three consecutive drug sessions, the three-cue discrimination procedure was initiated. Subjects received 38 sessions of preliminary drug discrimination training, of which 20 were preceded by the 4.0-mg/kg training dose of CDP.

Three-cue discrimination training. Within each block of 30 sessions, the 8.0-mg/kg dose of CDP was administered before nine sessions, one of the seven lower training doses of CDP (4.0, 2.8, 2.0, 1.4, 1.0, 0.7, or 0.5 mg/kg) was administered before nine sessions, and saline was administered before 12 sessions. Within each block of 30 sessions, only one of the seven lower training doses was given, and this dose was decreased across blocks. The sequence of injections given during each block of 30 sessions was: 8, S, L, L, S, 8, S, L, 8, S, L, S, 8, S, 8, L, S, L, S, 8, L, S, 8, S, L, 8, S, 8, L, S, where 8 = 8 mg/kg CDP, L = lower training dose of CDP, and S = saline. The same sequence of injections was used for all subjects, who each received a total of 210 drug discrimination training sessions during the three-cue discrimination training phase.

Three-cue discrimination retraining. To assess the effects of the preceding training on discriminative control by a wider range of training doses, three of the lower training doses (2.0, 1.4, and 1.0 mg/kg CDP) were retrained. Ninety sessions of drug discrimination training were conducted during retraining, and the training procedures used were identical to those described earlier.

Drugs

Chlordiazepoxide hydrochloride was generously donated by Hoffmann-LaRoche Inc. (Nutley, NJ) and was dissolved in 0.9% saline to a volume equivalent to 1.0 ml/mg. All drug doses refer to the total salt.

Data Analysis

All analyses were performed on percentage drug-appropriate choices on the first trial of each session. The effects of training blocks were assessed by one-way repeated-measures analysis of variance (ANOVA) using SAS-GLM procedure. Interactions between phase and training blocks were assessed by two-way repeated-measures ANOVA using SAS-GLM procedure. Tukey's HSD posthoc comparisons, using an α level of 0.05, provided comparisons between individual points.

RESULTS

Mean percentage drug-appropriate choices during the three-cue training phase for 8.0-mg/kg training dose sessions, lower training dose sessions, and saline sessions were plotted as a function of training dose blocks (Fig. 1). Mean percentage drug-appropriate choices during training for 8.0-mg/kg training dose sessions were 94.47% and were > 85% for each of the seven blocks of training doses. One-way repeated-measures ANOVA revealed that the mean percentage drug-appropriate choices for the 8.0-mg/kg training dose sessions did not differ across these seven blocks of training: $F(6, 66) < 1$. Mean percentage drug-appropriate choices during training for saline sessions were 11.53% and were < 20% during each of the seven blocks of training doses. One-way repeated-measures ANOVA revealed that mean percentage drug-appropriate choices for saline training sessions did not differ across these seven blocks of training: $F(6, 66) < 1$. Mean percentage drug-appropriate choices were directly related to training dose, with the three lowest training doses (1.0, 0.7, and 0.5 mg/kg CDP) producing predominantly saline-appropriate choices. The training dose correctly identified on the first trial on 50% of the training dose sessions was 1.4 mg/kg.

Mean percentage drug-appropriate choices during retraining for the 8.0-mg/kg training dose sessions, lower training

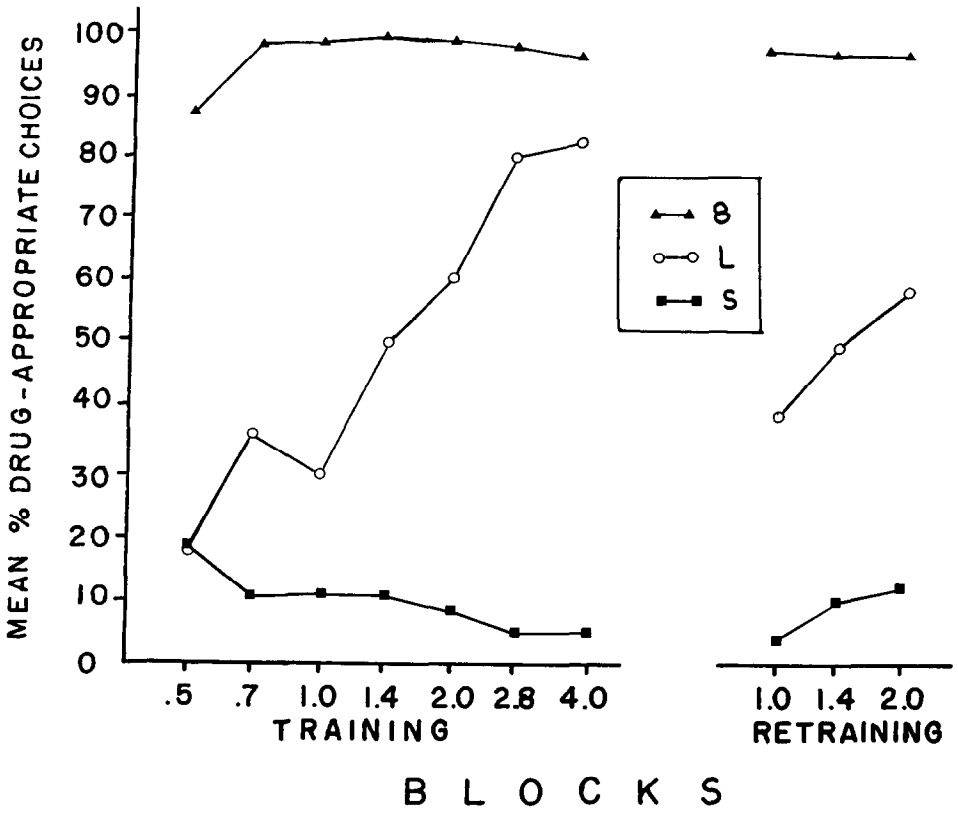

FIG. 1. (Left panel). Mean percentage drug-appropriate choices as a function of type of presession injection during each of the seven blocks of the training phase. The seven lower training doses were 4.0, 2.8, 2.0, 1.4, 1.0, 0.7, and 0.5 mg/kg CDP. (Right panel). Mean percentage drug-appropriate choices as a function of type of presession injection during each of the three blocks of the retraining phase. The three lower training doses were 2.0, 1.4, and 1.0 mg/kg CDP. Y-axis: mean percentage drug-appropriate choices on the first trial of each session. X-axis: blocks of 30 sessions with each of the lower training doses plotted on a log (base 2) scale. Each of the blocks consisted of nine sessions with the 8.0-mg/kg training dose of CDP (8), nine sessions with one of the lower training doses of CDP (L), and 12 sessions with saline (S). The range of the SEM was 4.4–6.6.

dose sessions, and saline sessions were plotted as a function of the three training dose blocks in Fig. 1. Mean percentage drug-appropriate choices during retraining for 8.0-mg/kg training dose sessions were 97.47% and were at least 90% for each of the three training blocks. One-way repeated-measures ANOVA revealed that mean percentage drug-appropriate choices for 8.0-mg/kg training dose sessions did not differ across these three blocks of training: $F(2, 22) < 1$. Mean percentage drug-appropriate choices during retraining for saline sessions were 13.78% and were < 20% during each of the three training blocks. One-way repeated-measures ANOVA revealed that mean percentage drug-appropriate choices for saline training sessions did not differ across these three blocks of training: $F(2, 22) = 2.18$; $p > 0.20$. Mean percentage drug-appropriate choices during retraining were directly related to training dose, and the training dose correctly identified on the first trial on 50% of the training dose sessions was 1.4 mg/kg. This was the same as the training dose correctly identified on 50% of the sessions during earlier training.

Phase (training vs. retraining) and the three lower training doses common to both phases (2.0, 1.4, and 1.0 mg/kg CDP) were entered into a 2 × 3 repeated-measures ANOVA. The analysis revealed no significant effect of phase [$F(1, 11) < 1$], a significant effect of dose [$F(2, 22) = 26.95$, $p < 0.01$], and no significant interaction between phase and dose [$F(2, 22) = 1.97$, $p > 0.15$]. Effects of phase on each of the three lower training doses common to both phases were evaluated individually using Tukey's HSD post hoc comparisons ($\alpha = 0.05$). Each of three pairwise comparisons revealed that there were no significant differences in mean percentage drug-appropriate choices between the training and retraining phases for the 2.0, 1.4, and 1.0 mg/kg training doses.

Two separate two-way repeated-measures ANOVAs were performed on mean percentage of drug-appropriate choices from 8.0-mg/kg training dose sessions and saline sessions across the two phases. Each two-way repeated-measures ANOVA included training dose blocks (i.e., blocks during which the 2.0, 1.4, and 1.0 mg/kg training doses were administered) and phase (training vs. retraining) as factors. Two-way repeated-measures ANOVA performed on percentage drug-appropriate choices during 8.0-mg/kg dose sessions revealed no significant effects of either training dose blocks [$F(2, 22) < 1$] or phase [$F(1, 11) = 2.06$, $p > 0.05$]. A similar analysis performed on percentage drug-appropriate choices during saline sessions also revealed no significant effects of either training dose blocks [$F(2, 22) = 2.30$, $p > 0.05$] or phase [$F(1, 11) < 1$].

DISCUSSION

The results reveal that these drug discrimination training procedures produce discriminative control by the CDP cue that is maintained during and after training with doses that yield predominantly saline-appropriate choices. Maintenance of control during training with low doses is revealed during the training phase (left panel of Fig. 1) by the consistently high percentage of drug-appropriate choices on 8.0-mg/kg training dose sessions, as well as by the consistently low percentage of drug-appropriate choices on saline training sessions. Similar effects were observed during the retraining phase (right panel of Fig. 1), suggesting that discriminative control by the 8.0-mg/kg training drug cue and the saline cue were maintained during and after training with lower training doses.

Similar conclusions are supported by evaluating performance at other doses. Discriminative control by three of the lower training doses (2.0, 1.4, and 1.0 mg/kg) was unaffected (right panel of Fig. 1) by intervening training with doses that had yielded predominantly saline-appropriate choices (1.0, 0.7, and 0.5 mg/kg). In addition, during both the training and retraining phases, the 1.4 mg/kg training dose was correctly identified on the first trial in 50% of the training sessions with that dose. These results suggest that discriminative control by a range of training doses of CDP (8.0, 2.0, 1.4, and 1.0 mg/kg) as well as control by saline is not reliably altered by intervening training with saline-like training doses.

Previous investigators observed the deterioration of discriminative control by the training drug cue after training with doses of the training drug that yielded vehicle-appropriate choices (15,17). Although those studies differed from each other in a number of ways, they shared in common the practice of providing extended training with doses of the training drug that engendered mostly vehicle-appropriate choices. The drug discrimination procedures employed in the present study, on the other hand, did not provide extended training with only vehicle-like cues, and control by the drug cue was reliably maintained during and after these training procedures. Thus, at the level of the group mean, these procedures allowed for repeated assessments of discriminative control by relatively low drug doses. In addition, at the level of the individual subject, variability between-subjects during both training and retraining was low, and, for each of the 12 subjects, performance during retraining approximated that previously observed for that subject during training.

Group and individual subject sensitivity is interesting to drug discrimination researchers (10,12,16,20), particularly as they relate to issues such as tolerance (6,11,19) and withdrawal (2,6,24), where pharmacologic treatments have been related to changes in sensitivity to the drug cue. Although the procedures presented here differ from those of typical drug discrimination studies, and therefore are limited in their generalizability, these procedures did maintain, at the group and individual subject level, long-term and stable performance across repeated assessments. These properties may be useful in the analysis of issues related to changes in drug discrimination performance, particularly when repeated assessment of low training doses is desirable.

ACKNOWLEDGEMENTS

This study was supported in part by a Charles and Johanna Busch Bequest Grant awarded to A.T.

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