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The Effects of Drug Discrimination History on Drug Discrimination and on Punished and Unpunished Responding

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LI, M. AND D. E. MCMILLAN. *The effects of drug discrimination history on drug discrimination and on punished and unpunished responding.* PHARMACOL BIOCHEM BEHAV **61**(1) 93–105, 1998.—Pigeons trained to discriminate methamphetamine from saline after a history of training to discriminate pentobarbital from saline responded on the drug key after both pentobarbital and methamphetamine, but the association of these drugs by reinforcing their discriminative stimulus responses on the same key did not influence their effects on either punished or unpunished responding. Similarly, pigeons trained to discriminate morphine from saline after a history of discrimination of buspirone from saline, responded on the drug key after both buspirone and morphine, but the association of their discriminative stimulus responses did not influence their effects on either punished or unpunished responding. Whether the effects of these drugs as discriminative stimuli, or their effects on punished and unpunished responding were studied first in the session did not influence the effects of these drugs. Low doses of methamphetamine or pentobarbital did not produce responding on the drug key in birds trained to discriminate higher doses of both drugs, but combinations of these doses did produce responding on the drug key. However, these combinations of low doses of methamphetamine with pentobarbital did not increase the rate-increasing effects of pentobarbital on punished responding. © 1998 Elsevier Science Inc.

Drug discrimination Behavioral history Punishment Methamphetamine Buspirone Morphine Phencyclidine Diazepam Cocaine Pigeons

WE have shown previously that pigeons can learn a series of drug discriminations using different training drugs as the discriminative stimuli (11). In these experiments pigeons were trained to discriminate pentobarbital from saline using a two-key procedure where responses on one key were reinforced if pentobarbital had been administered before the session, and responses on the other key were reinforced if saline had been administered before the session. After the discrimination had been established and a number of other drugs had been tested to determine if they substituted for the pentobarbital stimulus, the birds were divided into two smaller groups with methamphetamine replacing pentobarbital as the training drug in one group and morphine replacing pentobarbital as the training drug in the other group. Methamphetamine and morphine were quickly established as new discriminative stimuli. In drug substitution tests, animals responded on the key where responses were reinforced after methamphetamine administration, not only after the administration of methamphetamine and cocaine, but also after the administration of pentobarbital and diazepam, despite not having been exposed to pentobarbital as a training drug for several months. Similarly, in the other group, pigeons responded on the key where responses were reinforced after morphine administration not only after morphine, but also after pentobarbital and diazepam. These sequential discriminations were then extended to a third drug in both groups of birds. As each new training drug was added, not only was a new drug discrimination established, but the birds appeared to "remember" the previous drug discriminations even though they had not had additional training sessions with these drugs and training with other drugs as discriminative stimuli had intervened since the original training.

In the experiments described above, the pigeons had been reinforced for responding on the same key following the administration of two different drugs, each of which had been

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established as a discriminative stimulus for responding on that key. It is possible that the discriminative stimuli produced by these drugs might have become associated because the responses to these stimuli were being made on the same key. If the discriminative stimuli for the two drugs had become associated, it is possible that this association might influence other effects of these drugs on behavior to become more similar.

To test this possibility, we chose punished responding. A number of drugs including barbiturates, benzodiazepines, and buspirone increase rates of punished responding suppressed by response-produced electric shock (2,3,8,13). If a drug that increases punished responding is established as a discriminative stimulus, and then a second drug that does not increase punished responding is established as a discriminative stimulus using the same response location for both drugs, it is possible that the second drug might come to increase punished responding through the association of its discriminative stimulus properties with the discriminative stimulus properties of the first drug.

At the beginning of these experiments we had just completed a series of experiments where we had shown that the discriminative stimulus effects of drugs and their effects on other behaviors could be measured during the same experimental session (10). In these experiments, pigeons trained to discriminate pentobarbital from saline were also trained to respond under multiple variable-interval variable-interval schedules, where responses during one component of the multiple schedule were punished with electric shock (mult VI VI pun schedule). During test sessions, other doses of pentobarbital and other drugs were substituted for the training dose of pentobarbital to determine if they substituted for the training dose. Immediately after the short drug-discrimination session, the pigeons were tested under the mult VI VI pun schedule. In the drug discrimination experiments, increasing doses of pentobarbital produced increased responding on the drug key. When responding was measured under the mult VI VI pun schedule, pentobarbital also produced dose-dependent increases in the rate of punished responding, but not in the rate of unpunished responding. Diazepam produced effects similar to those of pentobarbital on both drug discrimination responding and punished responding, and to some degree so did phencyclidine. Buspirone did not substitute for pentobarbital as a discriminative stimulus, but it did increase punished responding. Methamphetamine neither substituted for pentobarbital as a discriminative stimulus, nor did it increase punished responding.

In the present experiments we used these same birds that had been used to study the discriminative stimulus effects of pentobarbital and the effects of pentobarbital on responding maintained under the mult VI VI pun schedule (10) and trained them to discriminate methamphetamine from saline instead of pentobarbital. Methamphetamine was chosen as the second training drug because the discriminative stimulus properties of pentobarbital do not generalize to amphetamines (12), nor do amphetamines increase punished responding in most experiments (8). Responses were reinforced on same key after methamphetamine administration where they had been reinforced after pentobarbital administration. After methamphetamine had been established as a discriminative stimulus, the effects of methamphetamine, pentobarbital, and other drugs were studied as discriminative stimuli and for their effects on responding maintained under the mult VI VI pun schedule to determine if the association of methamphetamine and pentobarbital as discriminative stimuli had caused their effects on punished responding to become similar.

To further extend our observations, a second group of pigeons were trained to discriminate buspirone from saline, and the effects of buspirone and other drugs were studied as discriminative stimuli and on responding maintained by the mult VI VI pun schedule. Buspirone was chosen as the training drug because it increases punished responding (13), but its discriminative stimulus properties do not generalize to pentobarbital (4). Subsequently, morphine replaced buspirone as the training drug in this group of birds and the effects of buspirone, morphine, and other drugs as discriminative stimuli and as drugs affecting responding under the mult VI VI pun schedule were redetermined. Morphine was chosen because it usually does not increase punished responding (8), nor are there reports in the literature that the buspirone discriminative stimulus generalizes to morphine.

METHODS

Subjects

Eight male White Carneux Pigeons (Palmetto Pigeon Plant, Sumter, SC), served as subjects in these experiments. Four of the birds had served previously to study the discriminative stimulus effects of pentobarbital and other drugs, as well as the effects of these same drugs on punished and unpunished responding (10). The other four birds were experimentally naive at the beginning of these experiments. All birds were individually housed with free access to food and water in a temperature- and humidity-controlled room that was maintained under a 12 L:12 D cycle (lights on at 0700 h). Birds were maintained at 80% (400–485 g) of their free-feeding weights prior to experimentation.

Apparatus

The experimental chamber was a Gerbrands Model G5610 (Gerbrands Corp., Arlington, MA) pigeon test cage equipped with three response keys, each of which could be transilluminated with several colors by a Gerbrands 28-V DC Key Light Assembly containing two 0.04-W bulbs for each color. A food hopper (Gerbrands) containing mixed grain was accessible to the pigeon when schedule contingencies were met. The chamber was enclosed inside a Gerbrands Model G7211 sound- and light-attenuating enclosure. A relay mounted inside the chamber operated whenever the key contacts were opened on the response key to provide auditory feedback for responses. A houselight illuminated the experimental chamber during the session except during a feed cycle when a light over the food hopper was illuminated. Electric shock (200 ms duration, 120 V AC, 60 Hz, mA individualized to the bird (range 1.0 to 3.5 mA; see Procedure) could be delivered through stainless steel electrodes implanted around each pubis bone (1). The electrodes were connected to the shock source by a plug attached to a leather harness which the bird wore at all times. The impedance of the electrodes was measured daily to ensure constancy of stimulus presentation. Schedule contingencies and data collection were programmed by a microcomputer (IBM 386 SX) through an interface (MED Associates, Inc., East Fairfield, VT). The microcomputer was housed in a room adjacent to the room containing the test chamber.

Procedure

Training. The methods for training pigeons have been discussed in detail previously (10,11). One group of birds had been trained previously to discriminate a 5.0 mg/kg dose of

pentobarbital from saline under a color-tracking procedure that used second-order reinforcement schedules (9). Briefly, a single peck on a white center key extinguished it and illuminated the two side keys, one with a red light and one with a green light. Five pecks on either key (fixed-ratio 5, or FR 5) darkened the side keys and lighted the center key to reinitiate the sequence. After each peck on the white center key, the position of the red and green key colors alternated randomly. After the completion of ten FR 5s on the "correct" side key, food was presented for 8 s. The red key was correct if 5.0 mg/kg pentobarbital had been administered 10 min before the session, and the green key was correct if saline had been administered 10 min before the session. This schedule was chosen because a large number of responses are required before the delivery of the first reinforcer (9).

After stimulus control was established for pentobarbital, the birds were trained to peck the center key under a multiple variable-interval 90-s variable-interval 90-s schedule where every fifth response under one of the two variable-interval schedules was punished with electric shock (mult VI VI pun schedule). The intensity of the electric shock was adjusted for each bird to reduce the rate of responding to about one-half the rate of unpunished responding. The key light was blue when responses were not punished and yellow when responses were punished.

After responding was stable under the VI VI pun schedule, training continued 6 days weekly with drug discrimination sessions occurring on 4 days (two saline and two pentobarbital sessions) and mult VI VI pun sessions occurring on 2 days. This training continued until the percentage of responses on the correct key was equal to or greater than 80% for six consecutive sessions, and the responding under the VI pun component of the mult schedule was occurring at about half the rate of responding occurring under the unpunished schedule component for each bird. When responding reached these criteria, animals were advanced to the test procedure.

In the present experiments, the birds continued the usual training (four drug discrimination sessions and two mult VI VI pun sessions weekly), except that 4.0 mg/kg methamphetamine was substituted for pentobarbital as the training drug. Once these birds met the same criteria for stability described above, they were exposed to test procedures again.

The second group of four birds was experimentally naive at the beginning of these experiments. These birds were trained to discriminate 3.0 mg/kg buspirone from saline using procedures identical to those described above. The only difference besides the use of buspirone as the first training drug was that the birds could meet the stability criterion by responding on the correct key 70% of the time, because drug discrimination was not as complete with buspirone as the training drug as it had been with pentobarbital. After drug discrimination stabilized, the pigeons were introduced to the mult VI VI pun schedule and later to both drug discrimination and the mult VI VI pun schedule as described above for pentobarbital. Once these birds met the criteria for stability, they also entered the testing procedure described below.

After completion of the test procedure with other drugs, 5.0 mg/kg morphine replaced buspirone as the training drug. Drug discrimination with morphine was established as described above for methamphetamine, and training sessions proceeded with four drug-discrimination training sessions and two mult VI VI pun sessions occurring weekly. After stability criteria were met, these birds entered the testing procedures again.

Test procedure. Test sessions consisted of two phases. In the first phase of the session drug-discrimination dose–response curves were determined to determine the generalization of the training dose (pentobarbital and later methamphetamine in the first group of birds; buspirone and later morphine in the second group of birds) to other doses of the training drug and to doses of other drugs. In the second phase of the session, the effects of the same drugs and doses on punished and unpunished responding were determined. Briefly, a bird was injected intramuscularly and placed into the chamber. After a 10-min presession, the first part of the session was initiated, and it was terminated by delivery of the first food reinforcer, which occurred whenever the bird completed ten FR 5s on either key. Immediately after food delivery the second part of the session began with the punishment component of the mult VI VI pun schedule. The VI VI pun schedule was in effect for 30 min, with each component presented for 15 min. These test procedures with drugs were conducted on Fridays with Thursdays serving as vehicle control sessions that were used to estimate baseline variability. Drug discrimination training, or training under the mult VI VI schedule continued on 4 other days of the week.

In these experiments, the effects of drugs on drug discrimination always were studied before the effects of drugs on responding under the mult VI VI pun schedule. To determine if the order of schedule presentation was an important independent variable, after the completion of these experiments in the first group of pigeons trained to discriminate methamphetamine from saline after a history of training to discriminate pentobarbital from saline, the experiments were repeated with drug effects on responding under the mult VI VI pun schedule determined prior to the effects of drugs on drug discrimination on test days. The pigeons were given drugs or saline solution and testing began 5 min later with the pun component of the multiple VI VI pun schedule presented for 10 min, followed by the nonpunishment component for 10 min. Immediately thereafter, the drug-discrimination phase of the test began. When the effects of pentobarbital as a discriminative stimulus appeared to be influenced by whether the drug discrimination experiments preceded or followed the experiments on punished and unpunished responding, the birds received several additional weeks of drug discrimination training with 5.0 mg/kg pentobarbital (the original training drug) replacing methamphetamine as the training drug. Subsequently, the effects of pentobarbital as a discriminative stimulus and its effects on punished and unpunished responding were determined again with the experiments on responding under the mult VI VI pun schedule preceding those on drug discrimination during test days. Additional experiments to study the effects of other drugs were not conducted using this procedure.

Data Analysis

Data obtained from the drug discrimination phase of the test sessions were plotted as a percentage of responses on the red key (hereafter referred to as the drug key). The mean response rate for responding on the two side keys was also determined and presented in tabular form. Data from the second phase of the session under the mult VI VI pun schedule were averaged to determine the effects of the drugs on punished and unpunished responding. The response rates during test sessions were compared with those after administration of the drug vehicle (Thursday sessions). Drug effects falling more than two standard deviations from these Thursday control means were considered to be statistically significant.

Drugs

Methamphetamine hydrochloride (Sigma Chemical Co., St. Louis, MO), pentobarbital sodium (Sigma Chemical Co.), phencyclidine hydrochloride (National Institute on Drug Abuse, Rockville, MD), cocaine hydrochloride (Sigma Chemical Co.), buspirone hydrochloride (Sigma Chemical Co.), and morphine sulfate (Mallinkrodt Chemical Co., St. Louis, MO) were dissolved in 0.9% physiological saline to concentrations allowing a volume of 1 ml/kg and administered intramuscularly into a breast muscle. Diazepam (Hoffmann–La Roche, Inc., Nutley, NJ) was dissolved in a solvent mixture (40% propylene glycol, 10% ethanol, and 50% saline solutions) and administered by the same route as the other drugs. Physiological saline and the diazepam solvent mixture were used for control injections. Successive injections were on alternate sides of the pigeons breast. Doses, except for diazepam, are expressed in mg/kg as the salt. Single observations were made at each dose in each animal.

RESULTS

Figure 1 shows the effects of drugs as discriminative stimuli in pigeons trained to discriminate pentobarbital from saline [top frame, data replotted from McMillan et al., 1997 (10)] and in pigeons trained to discriminate methamphetamine from saline after this history of pentobarbital discrimination (bottom frame). In pigeons trained to discriminate 5.0 mg/kg pentobarbital from saline, increasing doses of pentobarbital and diazepam produced an increasing percentage of responses on the drug key. Higher doses of both drugs produced as high a percentage of responses on the drug key as occurred during sessions when the training dose of pentobarbital was administered. Phencyclidine produced some responding on the drug key, but only about one-half as much as pentobarbital and diazepam. Methamphetamine and buspirone produced responding that was largely confined to the saline key.

The lower frame of Fig. 1 shows data for the effects of drugs on the percentage of responses on the drug key, after 4.0 mg/kg methamphetamine replaced pentobarbital as the training drug. Increasing doses of pentobarbital, methamphetamine, cocaine, and phencyclidine all produced increasing amounts of responding on the drug key with increasing dose. There was a slight dip in the methamphetamine dose–response curve after the 5.6 mg/kg dose of methamphetamine. After buspirone, responding was confined largely to the saline key.

Table 1 shows the effects of these drugs on overall rate of responding during the drug discrimination experiments. In birds trained to discriminate pentobarbital from saline, doses of 0.3 mg/kg buspirone and 0.1 and 0.3 mg/kg diazepam produced small increases (15–30% above the saline control mean) in the overall rate of responding. Higher doses of all drugs except pentobarbital decreased the overall rate of responding.

Table 1 also shows the effects of drugs on overall rates of drug-discrimination responding after methamphetamine replaced pentobarbital as the training drug. Only the 0.3 mg/kg dose of methamphetamine and the 3.0 mg/kg dose of pentobarbital increased the rate of responding. Higher doses of all drugs decreased rates of responding. These data show that dose levels up to those that were beginning to suppress overall rates of drug-discrimination responding were studied in these experiments.

Figure 2 shows the effects of these same drugs on punished responding determined during the same test sessions after completion of the drug-discrimination tests. In the pigeons

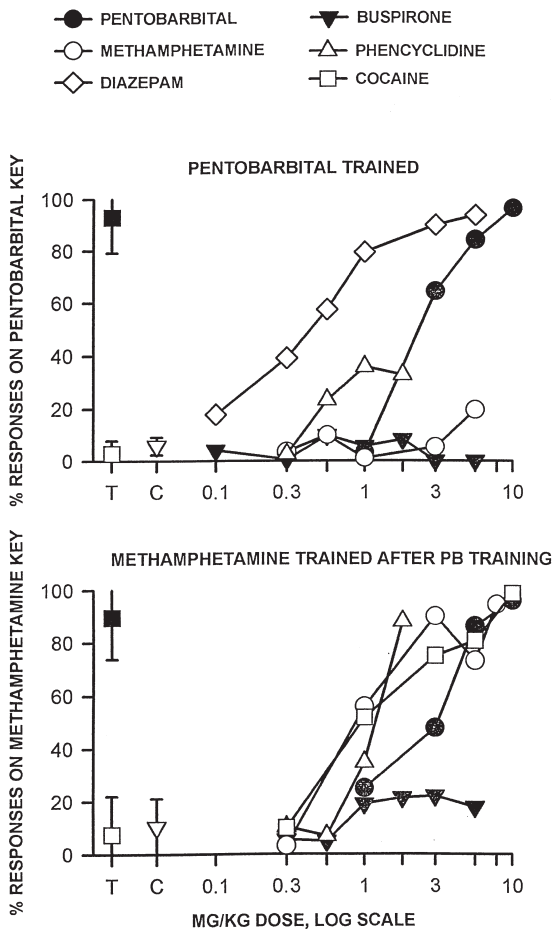

FIG. 1. Effects of drugs on drug-discrimination responding in pigeons trained to discriminate pentobarbital from saline (top frame) and in the same pigeons after methamphetamine had replaced pentobarbital as the training drug (bottom frame). Abscissa: dose, log scale. Ordinate: percentage of responses on the drug key (pentobarbital key in the top frame and methamphetamine key in the bottom frame). Points at T show the mean percentage of responses on the drug key ±2 standard deviations during pentobarbital, or methamphetamine training sessions (filled squares), or during saline training sessions (unfilled squares). Points at C show the mean ±2 standard deviations for saline control sessions run under the same procedures as the drug substitution tests. Each point on the dose–effect curves represent means of single observations in each of four birds.

trained to discriminate pentobarbital from saline (top frame), pentobarbital, diazepam, buspirone, and phencyclidine all increased punished responding. Methamphetamine did not increase punished responding. Pentobarbital and diazepam produced larger increases in punished responding (397 and 361% of the saline control mean) than did buspirone and phencyclidine (233 and 235% of the saline control mean).

The bottom frame in Fig. 2 shows the effects of drugs on punished responding after methamphetamine had replaced pentobarbital as the training drug. Pentobarbital continued to produce large increases in punished responding, and buspirone and phencyclidine continued to produce smaller increases in punished responding. Neither methamphetamine nor cocaine increased punished responding.

The effects of these drugs on responding during the VI component of the mult VI VI pun schedule where responses were not punished are shown in Table 2. When pentobarbital was the training drug, small increases in the rate of unpunished responding were produced by 3.0 mg/kg pentobarbital, 0.56 mg/kg buspirone, and 0.3 to 1.0 mg/kg phencyclidine. The increases in unpunished responding were small (about 120% of the saline control mean), compared to the very large in-

TABLE 1

EFFECT OF DRUGS ON DD RESPONDING RATE (RESPONSES/S) UNDER DIFFERENT TRAINING CONDITIONS

	Drugs						
Dose (mg/kg)	Pentobarbital	Busipirone	Methamphetamine	PCP	Diazepam	Cocaine	Morphine
Pentobarbital trained							
Control							
(Mean ± 2 SDs)	2.53 ± 0.98	2.35 ± 0.54	2.51 ± 1.16	2.72 ± 0.64	2.72 ± 0.3		
0.1	—	2.85	—	—	3.04↑	NT	NT
0.3	—	3.02↑	2.25	2.94	3.17↑	NT	NT
0.56	—	2.64	2.56	2.44	2.85	NT	NT
1.0	3.50	2.23	2.02	1.83↓	2.86	NT	NT
1.8	—	2.16	—	1.15↓	—	NT	NT
2.2	—	—	—	0.38↓	—	NT	NT
3.0	2.74	2.57	1.62	—	1.95↓	NT	NT
5.6	2.98	1.62↓	1.08↓	—	1.46↓	NT	NT
10.0	2.25	—	—	—	—	NT	NT
Methamphetamine trained after pentobarbital training							
Control							
(Mean ± 2 SDs)	2.7 ± 0.3	2.29 ± 0.42	2.58 ± 0.3	2.54 ± 0.56		2.79 ± 0.34	
0.3	—	2.52	3.23↑	2.08	NT	2.53	NT
0.56	—	2.17	—	2.23	NT	—	NT
1.0	1.97↓	1.86	2.55	2.06	NT	1.72↓	NT
1.8	—	1.69↓	—	1.23↓	NT	—	NT
2.2	—	—	—	0.42↓	NT	—	NT
3.0	3.02↑	1.38↓	1.91↓	—	NT	2.49	NT
5.6	2.57	1.43↓	1.48↓	—	NT	1.77↓	NT
7.8	—	—	1.25↓	—	NT	—	NT
10.0	2.32↓	—	—	—	NT	0.91↓	NT
Busipirone trained							
Control							
(Mean ± 2 SDs)	3.03 ± 0.6	3.32 ± 0.19	2.97 ± 0.22	3.38 ± 0.53	3.25 ± 0.18		
0.1	—	3.24	—	—	3.50↑	NT	NT
0.3	—	1.85↓	3.00	3.43	3.44	NT	NT
0.56	—	—	—	3.21	—	NT	NT
1.0	3.40	1.92↓	3.54↑	2.99	3.12	NT	NT
1.8	—	—	—	1.22↓	—	NT	NT
3.0	3.55	1.09↓	2.02	—	2.95↓	NT	NT
5.6	3.15	0.22↓	1.14↓	—	2.81↓	NT	NT
10.0	2.18	—	—	—	—	NT	NT
Morphine trained after busipirone training							
Control							
(Mean ± 2 SDs)	2.45 ± 0.24	2.45 ± 0.24	2.45 ± 0.24	2.45 ± 0.24	2.41 ± 1.16		2.45 ± 0.24
0.1	—	2.69	—	—	3.10	NT	—
0.3	—	2.62	2.48	1.92↓	2.86	NT	—
0.56	—	—	—	2.26	—	NT	—
1.0	2.77↑	2.82↑	1.89↓	1.97↓	2.27	NT	2.28
1.8	—	—	0.45↓	1.06↓	—	NT	—
3.0	2.89↑	1.55↓	0.32↓	—	2.10	NT	1.71↓
5.6	2.47	1.32↓	—	—	2.23	NT	2.07↓
10.0	1.68↓	—	—	—	—	NT	1.19↓
13.0	1.39↓	—	—	—	—	NT	—

↓ = Drug effect was more than 2 standard deviations below the control mean and ↑ = the drug effect was more than 2 standard deviations above the control mean. NT = the drug was not tested under the training condition. — = The dose was not tested in determining the dose-response curve.

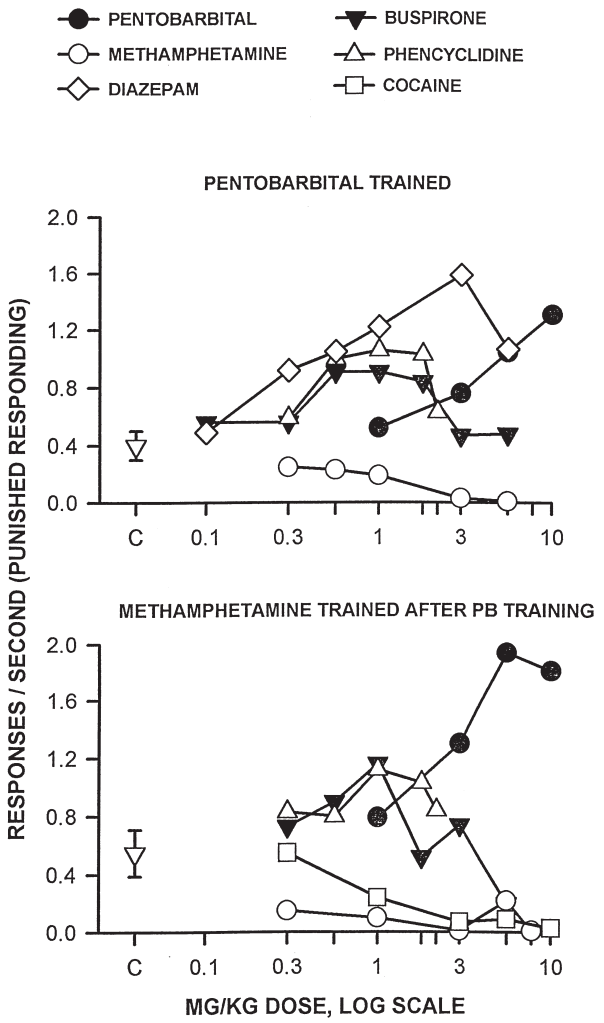

FIG. 2. Effects of drugs on punished and unpunished responding in pigeons trained to discriminate pentobarbital from saline (top frame) and in the same pigeons after methamphetamine had replaced pentobarbital as the training drug (bottom frame). Abscissa: dose, log scale. Ordinate: responses/s during the punishment component of the mult VI VI pun schedule. Points at C show the mean rate of punished responding ±2 standard deviations during control sessions when saline was administered. Each point on the dose–effect curves represents a mean of single observations in each of four birds.

creases in punished responding. No increases in unpunished responding occurred after diazepam. High doses of all of these drugs decreased rates of unpunished responding.

After methamphetamine replaced pentobarbital as the training drug in the drug discrimination experiments, doses of 3.0 mg/kg pentobarbital, 0.3 mg/kg methamphetamine, 0.56 and 1.0 mg/kg phencyclidine, and 3.0 and 5.6 mg/kg cocaine all produced small increases in unpunished responding. These increases were less than 20% above the saline control mean.

Figure 3 shows the effects of combinations of low doses of pentobarbital and methamphetamine as discriminative stimuli and on responding under the mult VI VI pun schedule in pigeons trained to discriminate methamphetamine from saline after a history of discrimination of pentobarbital from saline. The 1.0 and 3.0 mg/kg doses of pentobarbital resulted in responding largely on the saline key (top left frame). Doses of 0.3 to 3.0 mg/kg methamphetamine produced an increasing percentage of responses on the drug key with the 3.0 mg/kg dose producing more than 60% of the responses on the drug key. When 1.0 mg/kg pentobarbital was combined with increasing doses of methamphetamine, all doses of methamphetamine produced more responding on the drug key than had occurred when methamphetamine was given alone. A combination of 3.0 mg/kg pentobarbital with increasing doses of methamphetamine also produced a greater percentage of responses on the drug key than was obtained with the same doses of methamphetamine given alone.

Figure 3 also shows the effects of these combinations of pentobarbital and methamphetamine on overall rate of responding during the drug-discrimination part of the session, and on the rates of punished and unpunished responding during the second part of the session. None of the doses of pentobarbital or methamphetamine produced significant changes in overall rate of drug-discrimination responding (lower left frame). Combination of 1 mg/kg doses of pentobarbital and 1.0 and 3.0 mg/kg methamphetamine produced small but statistically significant decreases in rate of responding. Both doses of pentobarbital increased the rate of punished responding (upper right frame), while methamphetamine only decreased rates of punished responding. Methamphetamine blocked most of the increases in punished responding produced by pentobarbital. Neither pentobarbital nor methamphetamine, given alone or in combination, produced significant changes in the rate of unpunished responding (lower right frame).

Figure 4 shows the effects of drugs as discriminative stimuli in the second group of pigeons trained to discriminate 3.0 mg/kg buspirone from saline (top frame). Increasing doses of buspirone produced an increasing percentage of responses on the drug key. The two highest doses of buspirone produced more responding on the drug key than had occurred during training sessions. Pentobarbital, diazepam, phencyclidine, and methamphetamine produced responding that was largely confined to the saline key, although at some doses of all of the drugs except methamphetamine, the percentage of responses on the saline key slightly exceeded the percentages that occurred after saline administration during training sessions.

Figure 4 also shows the effects of drugs as discriminative stimuli in these pigeons after training with 5.0 mg/kg morphine substituted for buspirone as the training drug (bottom frame). The higher doses of morphine (5.6 and 10 mg/kg) and buspirone (3 and 5.6 mg/kg) produced responding on the drug key at a rate equal to or higher than that during drug-training sessions. Methamphetamine, pentobarbital, diazepam, and phencyclidine all produced responding primarily on the saline key. After drug-discrimination training with morphine as the training drug, the 10 mg/kg dose of pentobarbital produced responding that was almost equally distributed on the two keys; however, after 13 mg/kg pentobarbital responding again occurred predominately on the saline key.

Table 1 shows the effects of drugs on overall rates of drug-discrimination responding in pigeons trained to discriminate buspirone from saline, before and after morphine replaced buspirone as the training drug. When buspirone was the training drug the 1.0 mg/kg dose of methamphetamine and the 0.1 mg/kg dose of diazepam produced small increases (less than 20%) in overall rates of drug discrimination responding. High doses of all of the drugs, except pentobarbital, decreased responding. After morphine replaced buspirone as the training drug, the 1.0 and 3.0 mg/kg doses of pentobarbital and the 1.0

mg/kg dose of buspirone produced similar small increases in the overall rate of drug-discrimination responding. Higher doses of all of the drugs except diazepam produced statistically significant decreases in rates of responding.

Figure 5 shows the effects of these drugs on responding during the punishment component of the mult VI VI pun schedule both before and after morphine replaced buspirone as the training drug in the drug discrimination studies. Buspi-

TABLE 2

EFFECT OF DRUGS ON UNPUNISHED RESPONDING RATE (RESPONSES/S) UNDER DIFFERENT DD TRAINING CONDITIONS

	Drugs						
Dose (mg/kg)	Pentobarbital	Busipirone	Methamphetamine	PCP	Diazepam	Cocaine	Morphine
Pentobarbital trained							
Control							
(Mean ± 2 SDs)	1.2 ± 0.18	1.46 ± 0.18	1.47 ± 0.18	1.42 ± 0.1	1.45 ± 0.24		
0.1	—	1.42	—	—	1.31	NT	NT
0.3	—	1.17	1.37	1.58↑	1.50	NT	NT
0.56	—	1.75↑	1.57	1.86↑	1.26	NT	NT
1.0	1.25	1.36	1.33	1.72↑	1.17↓	NT	NT
1.8	—	1.44	—	1.27↓	—	NT	NT
2.2	—	—	—	0.59↓	—	NT	NT
3.0	1.46↑	0.72↓	1.20↓	—	0.91↓	NT	NT
5.6	1.14	0.70↓	0.88↓	—	0.60↓	NT	NT
10.0	0.96↓	—	—	—	—	NT	NT
Methamphetamine trained after pentobarbital training							
Control							
(Mean ± 2 SDs)	1.66 ± 0.14	1.71 ± 0.32	1.74 ± 0.16	1.69 ± 0.44		1.86 ± 0.2	
0.3	—	1.17↓	2.08↑	2.13	NT	1.80	NT
0.56	—	1.91	—	2.51↑	NT	—	NT
1.0	1.55	1.92	1.46↓	2.93↑	NT	2.05	NT
1.8	—	0.84↓	—	1.12↓	NT	—	NT
2.2	—	—	—	1.02↓	NT	—	NT
3.0	1.89↑	0.73↓	1.48↓	—	NT	2.09↑	NT
5.6	1.76	0.58↓	1.52↓	—	NT	2.18↑	NT
7.8	—	—	1.15↓	—	NT	—	NT
10.0	1.09↓	—	—	—	NT	0.87↓	NT
Busipirone trained							
Control							
(Mean ± 2 SDs)	1.26 ± 0.18	1.2 ± 0.24	1.18 ± 0.1	1.3 ± 0.19	1.4 ± 0.08		
0.1	—	1.37	—	—	1.24↓	NT	NT
0.3	—	1.09	1.30↑	1.28	1.29↓	NT	NT
0.56	—	—	—	1.47	—	NT	NT
1.0	1.24	0.93↓	1.28	1.75↑	1.28↓	NT	NT
1.8	—	—	—	1.46	—	NT	NT
3.0	1.39	0.68↓	0.65↓	—	1.28↓	NT	NT
5.6	1.41	0.66↓	0.53↓	—	1.00↓	NT	NT
10.0	1.21	—	—	—	—	NT	NT
Morphine trained after busipirone training							
Control							
(Mean ± 2 SDs)	1.2 ± 0.06	1.2 ± 0.06	1.2 ± 0.06	1.2 ± 0.06	1.22 ± 0.18		1.2 ± 0.06
0.1	—	1.16	—	—	1.31	NT	—
0.3	—	0.96↓	1.17	1.26	1.21	NT	—
0.56	—	—	—	1.40	—	NT	—
1.0	1.38↑	1.20	0.79↓	1.40	1.21	NT	1.18
1.8	—	—	0.63↓	1.68↑	—	NT	—
3.0	1.16	0.81↓	0.04↓	—	1.09	NT	1.09↓
5.6	1.16	0.71↓	—	—	1.10	NT	0.88↓
10.0	0.96↓	—	—	—	—	NT	0.34↓
13.0	0.83↓	—	—	—	—	NT	—
17.0	0.19↓	—	—	—	—	NT	—

↓ = Drug effect was more than 2 standard deviations below the control mean and ↑ = the drug effect was more than 2 standard deviations above the control mean. NT = the drug was not tested under the DD training condition. — = The dose was not tested in determining the dose-response curve.

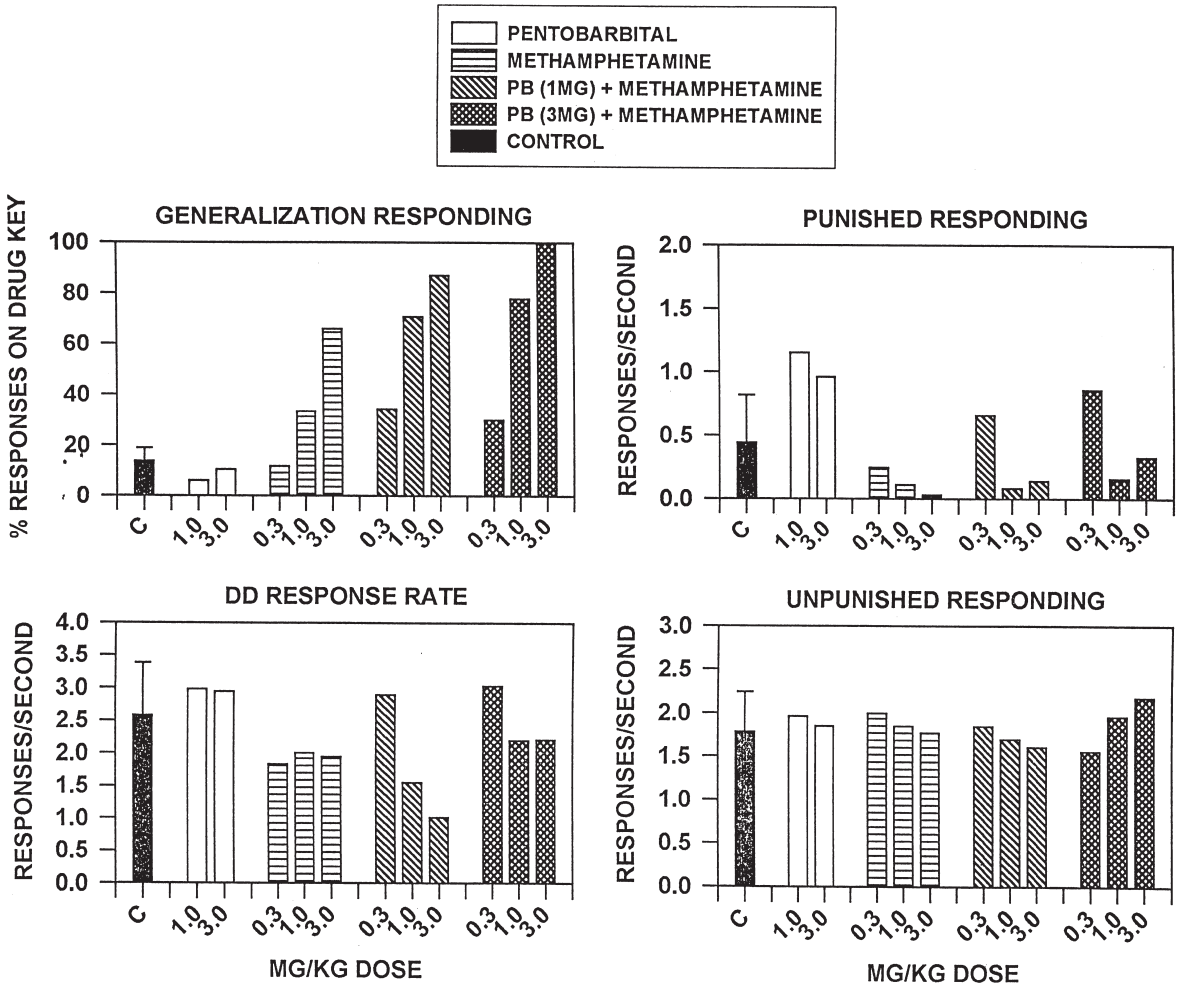

FIG. 3. Effects of pentobarbital, methamphetamine, and combinations of pentobarbital and methamphetamine on percentage of responding on the drug key (top left frame), overall rate of drug-discrimination responding (lower left frame), rate of punished responding (upper right frame), and rate of unpunished responding (lower right frame) in pigeons trained to discriminate methamphetamine from saline after a history of discriminating pentobarbital from saline. Abscissa: mg/kg dose. Ordinate: percentage of responses on the drug key (generalization responding), or rate of responding in responses/s. Each bar represents a mean of single observations in each of four pigeons. The bar at C shows the control means after saline administration. Brackets at C represent ±2 standard deviations around the control mean.

rone, pentobarbital, diazepam, and phencyclidine all increased rates of punished responding across a wide range of doses after buspirone discrimination training (top frame). The increases in punished responding were larger with pentobarbital and phencyclidine (312 and 332% of the saline control mean at the peak of the dose–effect curve) than with diazepam and buspirone (205 and 180% of the saline control mean at the peak of the dose–response curve). Methamphetamine failed to increase punished responding.

The effects of drugs on punished responding of birds trained to discriminate morphine after a history of buspirone discrimination are also shown in Fig. 5 (lower frame). Pentobarbital, diazepam, buspirone, and phencyclidine all increased punished responding, at one or more doses, although the increase in punished responding after buspirone occurred only after the 0.3 mg/kg dose and the increase was very small. At the peak of the dose–effect curve, pentobarbital and phencyclidine increased punished responding more (300 and 371% of the saline control mean) than did diazepam and buspirone (286 and 164%). Morphine and methamphetamine did not increase punished responding in these birds after morphine replaced buspirone as the training drug.

The effects of these drugs on unpunished responding during the VI component of the multiple schedule are shown in Table 2. When buspirone was the training drug, decreases in rates of unpunished responding were seen after buspirone (1.0 to 5.6 mg/kg), methamphetamine (3.0 and 5.6 mg/kg), and diazepam (all doses). The 0.3 mg/kg dose of methamphetamine and the 1.0 mg/kg dose of phencyclidine produced small increases in rates of unpunished responding. After morphine replaced buspirone as the training drug, the 1.0 mg/kg dose of pentobarbital and the 1.8 mg/kg dose of phencyclidine produced small increases in rates of unpunished responding (115 to 140% of the saline control mean), but none of the

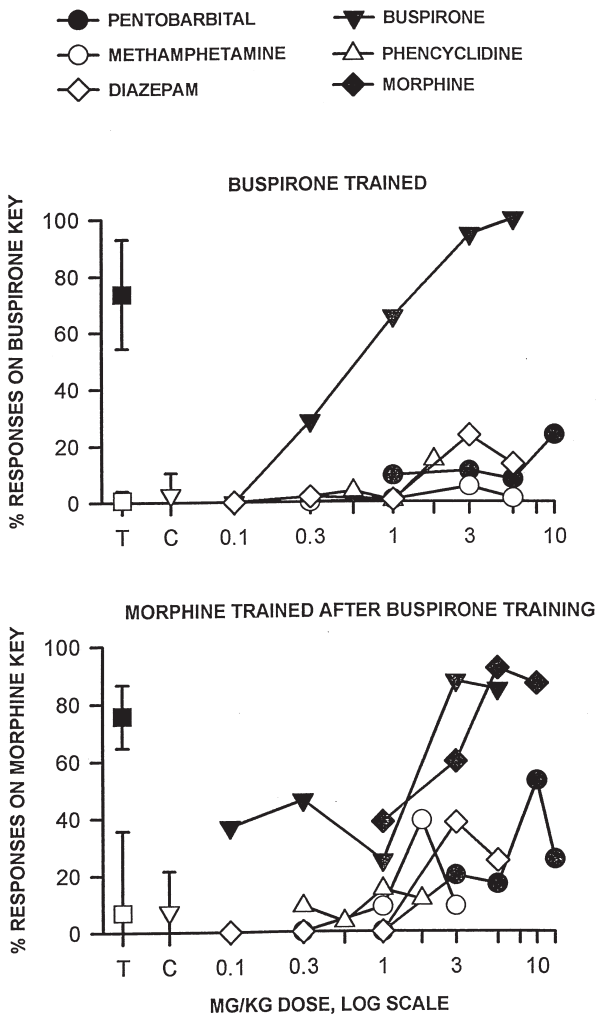

FIG. 4. Effects of drugs on drug-discrimination responding in pigeons trained to discriminate buspirone from saline (top frame) and in the same pigeons after morphine had replaced buspirone as the training drug (bottom frame). Abscissa: dose, log scale. Ordinate: percentage of responses on the drug key (buspirone key in the top frame and morphine key in the bottom frame). Points at T show the mean percentage of responses on the drug key ±2 standard deviations during buspirone, or morphine training sessions (filled squares), or during saline training sessions (unfilled squares). Points at C show the mean ±2 standard deviations for saline control sessions run under the same procedures as the drug substitution tests. Each point on the dose–effect curves represents a mean of single observations in each of four birds.

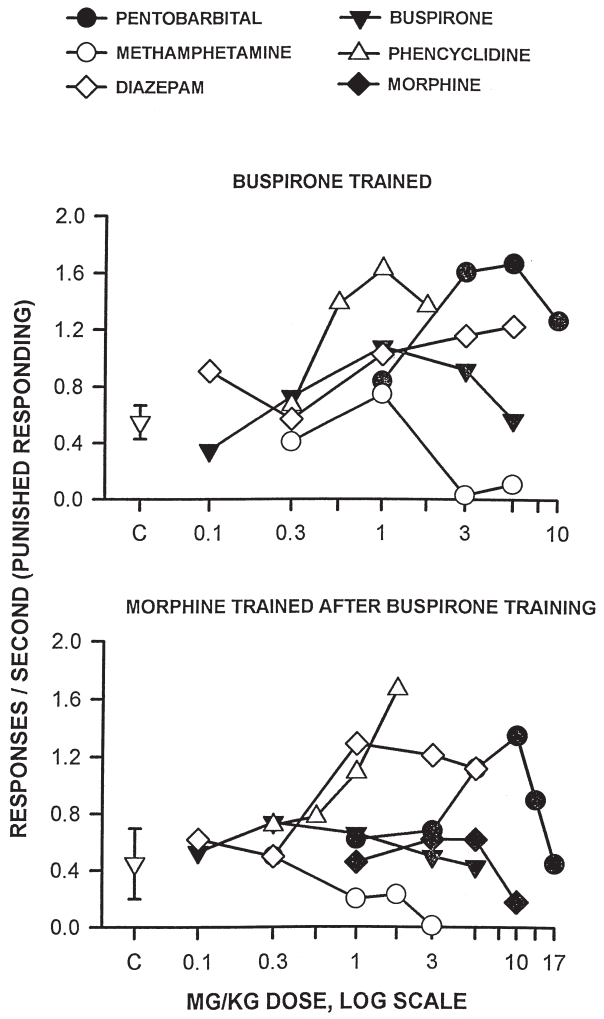

FIG. 5. Effects of drugs on punished responding in pigeons trained to discriminate buspirone from saline (top frame) and in the same pigeons after morphine had replaced buspirone as the training drug (bottom frame). Abscissa: dose, log scale. Ordinate: responses/s during the punishment component of the mult VI VI pun schedule. Points at C show the mean rate of punished responding ±2 standard deviations during control sessions when saline was administered. Each point on the dose–effect curves represents a mean of single observations in each of four birds.

other drugs produced significant increases in unpunished responding. Higher doses of all of the drugs except phencyclidine and diazepam decreased rates of unpunished responding.

Figure 6 compares the effects of drugs on drug discrimination responding when the order of test phases was reversed. These experiments were performed in the first group of pigeons trained to discriminate methamphetamine from saline after a history of training to discriminate pentobarbital from saline. The dose–response curves are shown when drug discrimination was studied before responding under the mult VI VI pun schedule (filled circles) and when drug discrimination was studied after responding under the mult VI VI pun schedule (unfilled circles). When pentobarbital discrimination was studied after punished and unpunished responding, the drug discrimination curve appeared to shift to the right, although the shift was not statistically significant. The dose–response curves for the discriminative stimulus effects of the other drugs did not shift.

Because the pentobarbital dose–response curve for drug discrimination appeared to shift to the right when drug discrimination was studied after the effects of pentobarbital on punished and unpunished responding were determined, to determine if this was related to a weakening of stimulus control by pentobarbital due to an extended period without further drug discrimination training with pentobarbital as the training drug, these birds were given several weeks of training with

pentobarbital reestablished as the training drug and then the pentobarbital dose–response curve was determined again. This dose–response curve (filled triangles) fell between the other two dose–response curves.

The comparison of the effects of these drugs on punished responding under the different orders of testing are shown in Fig. 7. Pentobarbital, buspirone, and PCP increased punished responding to the same extent, regardless of whether the ef-

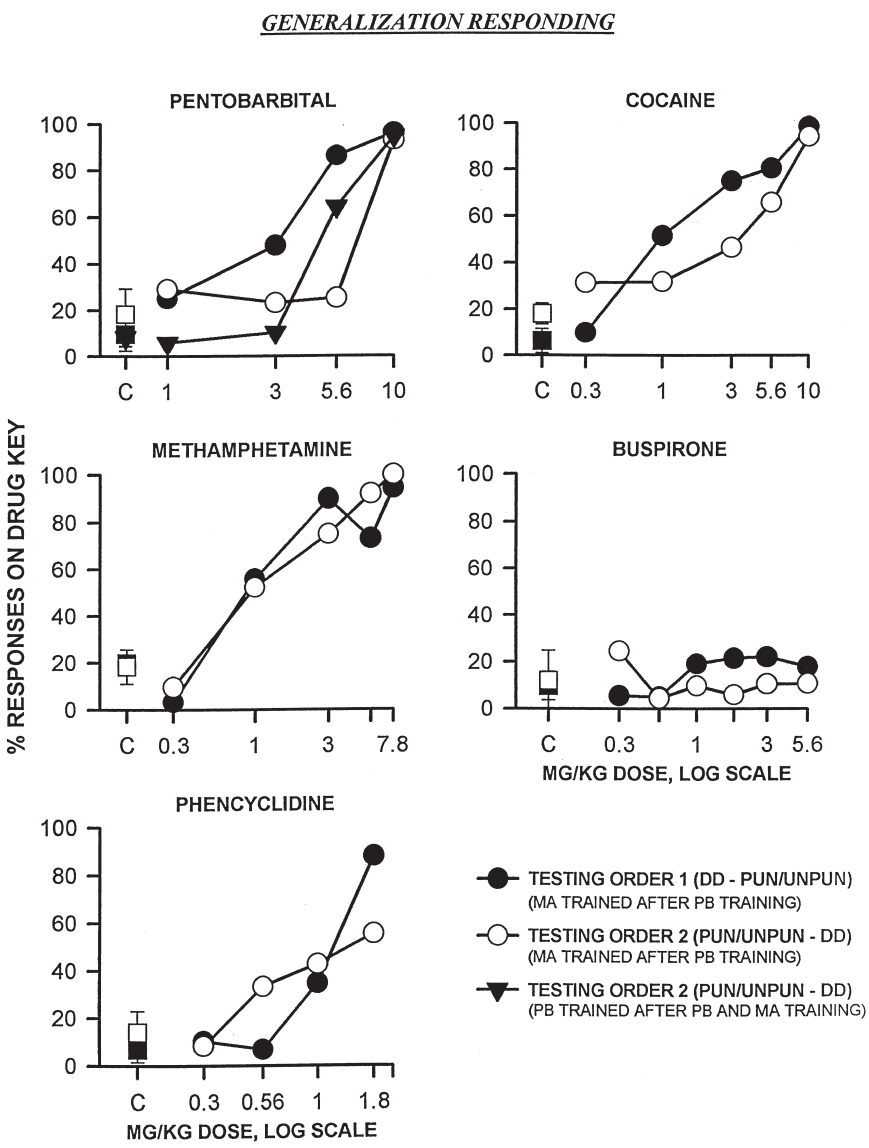

FIG. 6. Effects of drugs on drug-discrimination responding in pigeons trained to discriminate methamphetamine from saline after a history of discrimination of pentobarbital from saline. Pigeons were tested with the drug-discrimination phase conducted before the mult VI VI pun phase (filled points) and in the same pigeons with the drug discrimination phase conducted after the mult VI VI pun phase (unfilled points). The effects of pentobarbital were also studied after additional training with pentobarbital as the training drug with the drug discrimination phase conducted after the mult VI VI pun phase (filled triangles). Abscissa: dose, log scale. Ordinate: percentage of responses on the drug key. Points at C show the mean percentage of responses on the drug key ±2 standard deviations during sessions when saline was administered (filled squares when drug discrimination was studied before mult VI VI pun and unfilled squares, or filled triangles when drug discrimination was studied after mult VI VI pun). Each point on the dose–effect curves represents a mean of single observations in each of four birds.

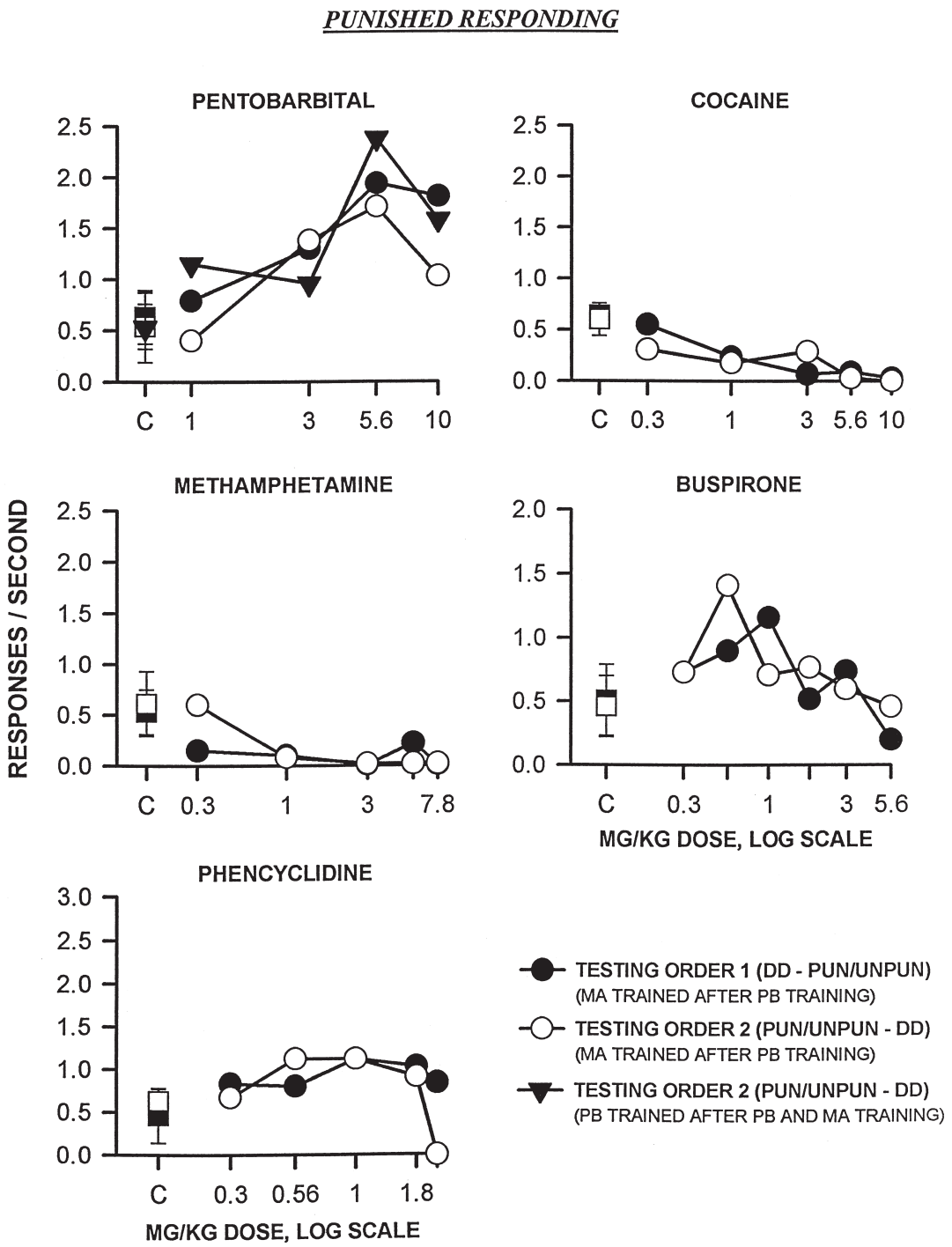

FIG. 7. Effects of drugs on punished responding in pigeons trained to discriminate methamphetamine from saline after a history of discrimination of pentobarbital from saline. Pigeons were tested with the drug-discrimination phase conducted before the mult VI VI pun phase (filled points) and in the same pigeons with the drug discrimination phase conducted after the mult VI VI pun phase (unfilled points). The effects of pentobarbital were also studied after additional training with pentobarbital as the training drug with the drug discrimination phase conducted after the mult VI VI pun phase (filled triangles). Abscissa: dose, log scale. Ordinate: rate of punished responding in responses/s. Points at C show the mean rate of punished responding ±2 standard deviations during sessions when saline was administered (filled squares when drug discrimination was studied before mult VI VI pun and unfilled squares, or filled triangles when drug discrimination was studied after mult VI VI pun). Each point on the dose–effect curves represents a mean of single observations in each of four birds.

fects of the drugs on punished responding were determined before or after the drug discrimination tests. Cocaine and methamphetamine did not increase punished responding. The dose–effect curves for all of these drugs were not different, regardless of the order in which the drugs were studied. There was a similar lack of difference in the dose–effect curves for overall rate of responding during drug-discrimination sessions and for effects of drugs on unpunished responding (data not shown).

DISCUSSION

The main purpose of these experiments was to determine if the reinforcement of drug-discrimination responses on the same response key following discrimination training with two drugs with different pharmacological properties would cause other effects of these drugs on behavior to become more similar. The answer to this question appears to be no, at least with respect to the drugs and behaviors studied in these experiments. In the first set of experiments, pigeons were trained to respond on one key after pentobarbital was administered and a different key after saline was administered. In these birds, other doses of pentobarbital and diazepam and to some extent PCP, generalized to the training dose of pentobarbital, but methamphetamine and buspirone did not. All of these drugs except methamphetamine increased punished responding. After methamphetamine replaced pentobarbital as the training drug under the drug-discrimination procedure, pigeons also responded on the drug key after methamphetamine administration. However, the association between responding on the same drug key following the administration of pentobarbital and methamphetamine did not cause methamphetamine to increase punished responding, nor did it alter the shape and position of the dose–response curves for the effects of any of the other drugs studied on either punished or unpunished responding to any great extent.

Pentobarbital is known to increase punished responding (5,8), while methamphetamine does not (5). In fact, amphetamines have sometimes been shown to further decrease responding suppressed by punishment. It was possible that the pharmacological effects of pentobarbital and methamphetamine on punished responding were too different for the association between their stimulus effects to influence their effects on punished responding. Therefore, the experiment was replicated in a new group of birds where buspirone was the first training drug in drug-discrimination experiments and morphine was the second training drug. Buspirone increases punished responding (13), and generally morphine does not (5,8), although there have been a few reports where morphine has been found to increase punished responding (6,7). In pigeons trained to discriminate buspirone from saline, generalization of the buspirone discriminative stimulus was not obtained for any of the other drugs that were studied, although a number of these drugs increased punished responding, including pentobarbital, diazepam, and phencyclidine.

When morphine replaced buspirone as the training drug, responding occurred on the drug key after both buspirone and morphine. Buspirone continued to increase punished responding before and after it was replaced as the training drug by morphine; however, the association of responding on the same drug key in the presence of the discriminative stimuli produced by buspirone and morphine did not result in increases in punished responding after morphine, nor did it systematically alter the shape or position of any of the dose–effect curves for the effects of these drugs on punished and unpunished responding. Thus, the association of the discriminative stimulus properties of two drugs by reinforcing responding on the same key after the administration of either drug does not appear to influence other pharmacological effects of these drugs.

This observation can be extended to drug interactions. We have shown previously that doses of morphine and methamphetamine that do not substitute for pentobarbital as discriminative stimuli, nor potentiate the discriminative stimulus effects of pentobarbital when given in combination with pentobarbital, do potentiate the discriminative stimulus effects of pentobarbital when animals have been trained to discriminate either morphine or methamphetamine after a history of pentobarbital discrimination (11). The present experiments replicate this interaction between pentobarbital and methamphetamine for drug discrimination. Combinations of pentobarbital and methamphetamine that produced little responding on the drug key when given alone, produced responding predominantly on the drug key when combined in the pigeons that had received sequential drug discrimination training with pentobarbital and methamphetamine. However, these interactions did not extend to other drug effects. There were no interactions between pentobarbital and methamphetamine for unpunished responding, and methamphetamine blocked the increases in punished responding produced by pentobarbital rather than potentiating these effects. This is further evidence that the association of two drugs as discriminative stimuli for the same response does not influence their other pharmacological effects.

Although we were not successful in demonstrating that associating the discriminative stimulus properties of two drugs by requiring responses to occur on the same key after both drugs could influence other pharmacological effects of these drugs, we did replicate a number of previous findings. First, we have shown previously that animals trained to discriminate drugs from saline can be used to measure the effects of other drugs on behavior during the same test sessions (10). The present experiments replicate these findings in two different groups of pigeons using two different training drugs.

Second, we have shown previously that pigeons can be trained to discriminate the presence or absence of two or more drugs, by reinforcing responses on the same key after either of these drugs (11). The present experiments provided further demonstrations of this phenominon, extending it to additional sequential drug training with buspirone and morphine. The implications of this finding for relapse to drug use have been discussed previously (11). In the present experiments, we combined these techniques to show that pigeons can be trained to respond on the same key following the administration of two or more drugs, and then can be used to study other drug effects on behavior immediately after the drug discrimination sessions. Not only does this replicate our previous findings, but it also provides a new tool for studying the relationship between the stimulus properties of drugs and their other pharmacological effects.

The only drug whose effects appeared to be altered greatly after the initial drug was replaced with a new training drug was phencyclidine. In pigeons trained to discriminate pentobarbital from saline, phencyclidine never generated more than 36% responding on the drug (pentobarbital) key. After the substitution of methamphetamine for pentobarbital as the training drug, 88% of the responses after the 1.7 mg/kg dose of phencyclidine occurred on the drug key. It is possible that the chronic administration of methamphetamine did influence phencyclidine discrimination. However, we have observed a

partial generalization of the pentobarbital stimulus to phencyclidine in previous experiments (10). The difference in the phencyclidine discrimination curve before and after methamphetamine replaced pentobarbital as the training drug may represent the variability in the degree to which the phencyclidine and pentobarbital stimuli can substitute for each other in drug discrimination experiments.

In previous experiments where the effects of drugs as discriminative stimuli and their effects on other behaviors were studied during the same session, the discriminative stimulus effects were always studied first. One of the purposes of the present study was to determine if the order in which the behaviors were studied was an important determinant of the drug effect. The answer to this question appears to be a qualified no. The effects of methamphetamine, PCP, cocaine, and buspirone did not depend on whether drug-discrimination responding or responding under the mult VI VI pun schedule was studied first. The same statement can be made for the effects of pentobarbital on both punished and unpunished responding. The pentobarbital dose–response curve for drug discrimination appeared to shift to the right when drug discrimination was studied after punished and unpunished responding rather than before, but the shift was not statistically significant.

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