Pharmacology Biochemistry and Behavior, Vol. 2, pp. 741–746. ANKHO International Inc., 1974. Printed in the U.S.A.

Repeated Acquisition of Response Sequences: Effects of *d*-Amphetamine and Chlorpromazine[1]

DONALD M. THOMPSON

Department of Pharmacology, Georgetown University, Schools of Medicine and Dentistry
Washington, D.C. 20007

(Received 10 January 1974)

THOMPSON, D. M. *Repeated acquisition of response sequences: effects of d-amphetamine and chlorpromazine.* PHARMAC. BIOCHEM. BEHAV. 2(6) 741–746, 1974. – Pigeons obtained food by making 4 responses on 3 keys in a specified sequence, e.g., left, right, center, right. All 3 keys were the same color throughout the response sequence. Under the learning condition, the four-response sequence was changed from session to session. After learning (within-session error reduction) had stabilized, this baseline of repeated acquisition was used to assess the effects of varying doses of *d*-amphetamine and chlorpromazine. For comparison, the drug tests were also conducted under a performance condition, in which the four-response sequence was the same from session to session. Increases in total errors and pausing were obtained at the largest dose of each drug under both the learning and performance conditions. Under the learning condition, the error rate decreased across trials within each session, but the degree of negative acceleration was less in the drug sessions than in the control sessions. In contrast, under the performance condition, the error rate was relatively constant across trials, but was higher in the drug sessions than in the control sessions.

Acquisition Performance Tandem response sequences *d*-Amphetamine Chlorpromazine Pigeons

A previous study from this laboratory used repeated acquisition of behavioral chains as a baseline to assess the effects of varying doses of *d*-amphetamine and chlorpromazine [25]. Pigeons worked for food in a chamber containing 3 response keys; all 3 keys were illuminated at the same time by one of four colors. For each session the pigeon's task was to learn a new four-response chain by pecking the correct key in the presence of each color, e.g., keys yellow – Left correct; keys green – Right correct; keys red – Center correct; keys white – Right correct; food. Drug administration (intramuscularly, 30 min presession, once a week) began after the total errors per session (overall accuracy) and the within-session error reduction (learning) had stabilized. The largest dose of *d*-amphetamine (4 mg/kg) was found to impair overall accuracy and to decrease the rate of learning; smaller doses either produced progressively less impairment or had no effect. In contrast, chlorpromazine did not affect overall accuracy at any of the doses tested (0.5–8 mg/kg), although there was a slight error-increasing effect at the largest dose during the first part of the session.

In the chain procedure described above, both color and serial position were available as discriminative stimuli for correct responding. In the present research, different colored keylights were no longer associated with the four-response sequence; when the keylights were on, they were always white. Such a situation, in which reinforcement is contingent upon the completion of four behavioral requirements in succession in the absence of correlated external stimuli, can be termed a "tandem" sequence (cf. [7]). In short, the tandem procedure eliminated color as a discriminative stimulus so that the pigeons had only serial position as a cue for pecking the correct keys, e.g., Left, Right, Center, Right, food.

The main objective of the present research was to determine whether the repeated acquisition of tandem response sequences would be affected by *d*-amphetamine and chlorpromazine in the same way that was found with behavioral chains in the previous study [25]. It has been shown in a variety of other situations that the behavioral effects of many drugs can be modified by the presence or absence of external discriminative stimuli [5, 10, 11, 15, 16, 17, 19, 20, 22, 24, 27, 28]. To permit a further comparison, the drug tests were also conducted under a performance condition, in which the tandem response sequence was the same from session to session.

[1] This research was supported by Public Health Service Grants MH 22340, FR 5360, and FR 5306. The *d*-amphetamine and chlorpromazine were kindly donated by Smith Kline and French Laboratories. I wish to thank Dr. Annette S. Thompson for comments on the manuscript.

METHOD

Animals

Three adult male White Carneaux pigeons (Nos. 7, 8 and 10) were used. All had been used previously in drug experiments involving the repeated acquisition and performance of behavioral chains [25,26]. The pigeons were maintained within 10 g of 80% of their free-feeding weights throughout the research by food presented during the sessions and by postsession supplemental feeding. The 80% values ranged between 475 and 510 g. Water and grit were always available in the home cages.

Apparatus

A standard three-key pigeon chamber (Lehigh Valley Electronics, Model 1519B) and connecting automatic control equipment were used. Each translucent response key, which required a static force of 18 g (0.177 newton) to close the microswitch, could be transilluminated by a Sylvania 24ESB white indicator lamp. The scheduling of events was accomplished by means of timers, steppers and associated relay circuitry; the recording was by counters and an 11 pen event recorder. White noise was continuously present in the chamber to mask extraneous sounds.

Procedure

Throughout the following procedures the reinforcer was 5 sec access to mixed grain. Presentation of food was accompanied by the offset of the keylights and the onset of the magazine light. The houselight was always off. Each session terminated after 40 food presentations. A blackout (all lights off) of variable duration preceded and followed each session. With few exceptions there were 7 daily sessions a week.

Baseline conditions. The pigeons obtained food by making 4 responses on 3 keys in a specified sequence, e.g., Left, Right, Center, Right (LRCR). All 3 keys were the same color (white) throughout the tandem sequence. (There was a momentary dimming of the keylights when the sequence advanced.) The same sequence (in this case, LRCR) was repeated throughout a given session and each completion of the sequence was considered a trial. Food reinforcement was on a fixed ratio (FR 5) schedule: the completion of every fifth trial was followed by 5 sec access to grain. The completion of all other trials was followed by a 0.5 sec presentation of the food magazine. The number of correct responses per session was fixed: four-response sequence on an FR 5 schedule for 40 food reinforcements = 800 correct responses. When the pigeon pecked an incorrect key (a key not included in the four-response sequence), the error was followed by a 5 sec timeout. During the timeout, the keylights were off and a response had no effect. An error did not reset the sequence; i.e., the correct key after the timeout was the same as before the timeout.

Under the tandem-learning condition, the four-response sequence was changed from session to session. The sequences were carefully selected to be equivalent in several ways and there were restrictions on their ordering across sessions (see [25]). An example of a typical set of six sequences is as follows: LRCR, CLRL, LRLC, RCRL, CLCR, RCLC.

Under the tandem-performance condition, the four-response sequence was the same from session to session. Different sequences were arbitrarily selected for the three pigeons: CRLR for No. 7; RCRL for No. 8; LRCR for No. 10.

Drug testing. The drugs were tested first under the tandem-learning condition and then under the tandem-performance condition with No. 7; the conditions were reversed with Nos. 8 and 10. Before the drug testing began, the tandem baseline (either learning or performance) was stabilized. The baseline was considered stable when the total errors per session and the within-session error rates no longer showed systematic change from session to session. Following baseline stabilization (50–70 sessions under the tandem-learning condition; 20–30 sessions under the tandem-performance condition), the next 16 weeks were used to obtain dose-effect data for *d*-amphetamine sulfate and chlorpromazine hydrochloride. Four doses of each drug were tested (0.5, 1, 2, and 4 mg/kg of *d*-amphetamine; 2, 4, 8, and 16 mg/kg of chlorpromazine) and two determinations for each dose were taken with each pigeon. The drug testing followed the design ACCA, where A and C represent the blocks of four doses of *d*-amphetamine and chlorpromazine; within each block, the doses were tested in a random order. The drugs were dissolved in saline and injected into the pectoral muscles 30 min before the test sessions, which took place once a week. Another session in each week was preceded by the administration of saline. The volume of each injection was 0.1 ml/100 g body weight.

RESULTS

Figure 1 shows the effects of varying doses of *d*-amphetamine and chlorpromazine (both determinations) on the total errors per session under the tandem-learning and tandem-performance conditions. The drug data for individual animals were analyzed by comparing a given drug session with the saline sessions and all of the baseline sessions during drug testing except the one after the drug session. The brackets indicate the ranges of variability for the baseline (B) and saline (S) sessions. A drug was considered to have an effect on overall accuracy to the extent that the dose data fell outside of both ranges (the two dashed horizontal lines). Note that the control error levels under the learning condition were higher and more variable than those under the performance condition. There were several consistencies in the drug data (both determinations) for the three pigeons: (1) under both the learning and performance conditions, both *d*-amphetamine and chlorpromazine impaired overall accuracy at the higher doses, (2) under both conditions, the error-increasing effect of *d*-amphetamine was obtained at lower doses than with chlorpromazine, and (3) with both drugs, the error-increasing effect was detected at lower doses under the performance condition than under the learning condition.

Figure 2 illustrates the within-session effects on accuracy obtained with the largest doses of *d*-amphetamine and chlorpromazine (first determinations) under the tandem-learning and tandem-performance conditions. The errors are plotted cumulatively so that the rate of errors during a given part of a session can be estimated easily from the slope of the curve. The curves for the drug sessions should be compared to the saline (max) and saline (min) sessions, which were the sessions with the maximum and minimum total errors of all the saline sessions (16) conducted during drug testing under a given condition. Under the learning condition, although the errors decreased across trials within each of the four sessions shown for each pigeon, the rate of

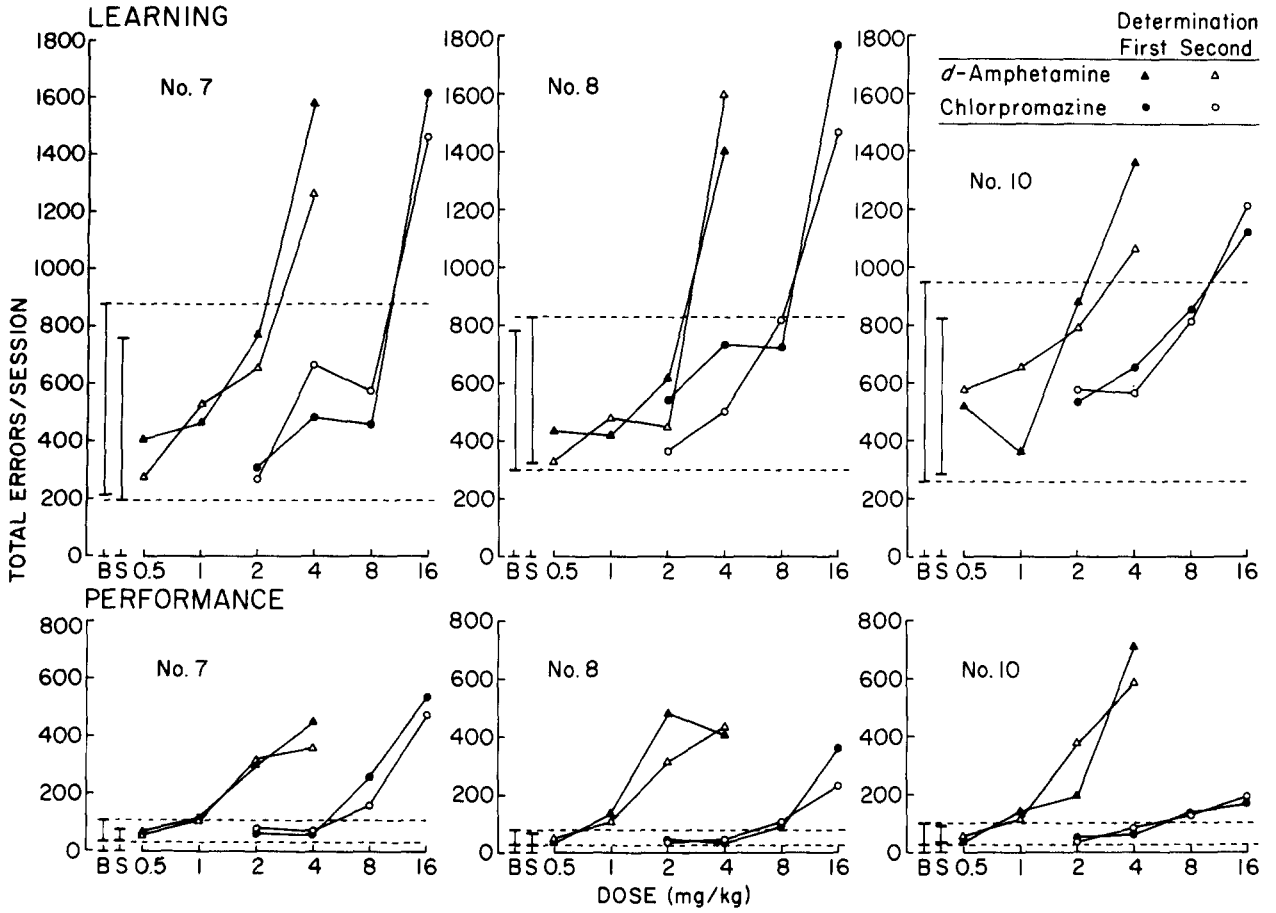

FIG. 1. Effects of *d*-amphetamine and chlorpromazine on the total errors per session under the tandem-learning and tandem-performance conditions. Four doses of each drug were tested and there were 2 determinations for each dose with each pigeon. The brackets and dashed horizontal lines indicate the ranges of variability for the baseline (B) and saline (S) sessions.

error reduction generally occurred more slowly during the drug sessions.

The change in error rate (negative acceleration) during each session under the learning condition (Fig. 2, top) was quantified by applying the Index of Curvature (cf. [8,25]) to the cumulative data. If all the errors occurred during the first 20 trials, the Index would take on its maximum value of −0.900; if the error rate were constant during the session, the Index would equal 0. The Index values are shown for each curve in Fig. 2. Note that under the learning condition, the degree of negative acceleration of error rate decreased (smaller Index values) as the total errors per session increased. Under the performance condition, however, the Index values were not consistently related to the total errors per session. In the control and drug sessions under the performance condition, there was either slight positive acceleration of error rate or less negative acceleration than that found under the learning condition. In short, the error rate under the performance condition was relatively constant, but was higher in the drug sessions than in the control sessions. The second determinations for these doses yielded similar results.

An inspection was made of the distribution of errors across the four serial positions of the tandem sequence and across the 5 serial positions of the fixed ratio schedule. The error distributions are not shown since there was no apparent drug effect. Under both the learning and performance control conditions, fewer errors tended to be made in the last parts of the tandem sequence and the fixed ratio schedule than in the first parts. The same trend was also detected throughout the testing of both drugs.

Although errors were the data of major interest, there were other behavioral measures affected by the drugs that should be mentioned. One of these was the total trial time (i.e., the total number of minutes that the keylights were on during a session), which indicates the amount of pausing that occurred. In general, the effects of the drugs on total trial time (not shown) were similar to their effects on total errors. When the drugs increased the total errors, the amount of pausing also generally increased. There were, however, a few cases where the total errors were above the control range but the total trial time was not, e.g., No. 8 at 4 mg/kg of *d*-amphetamine (first determination) under the learning condition; No. 10 at 2 mg/kg of *d*-amphetamine (both determinations) under the performance condition. There were no instances of increased pausing at doses that had no effect on accuracy.

Another behavioral measure affected by the drugs was the timeout responses per session, i.e., the total number of responses made during the 5 sec timeout periods when the

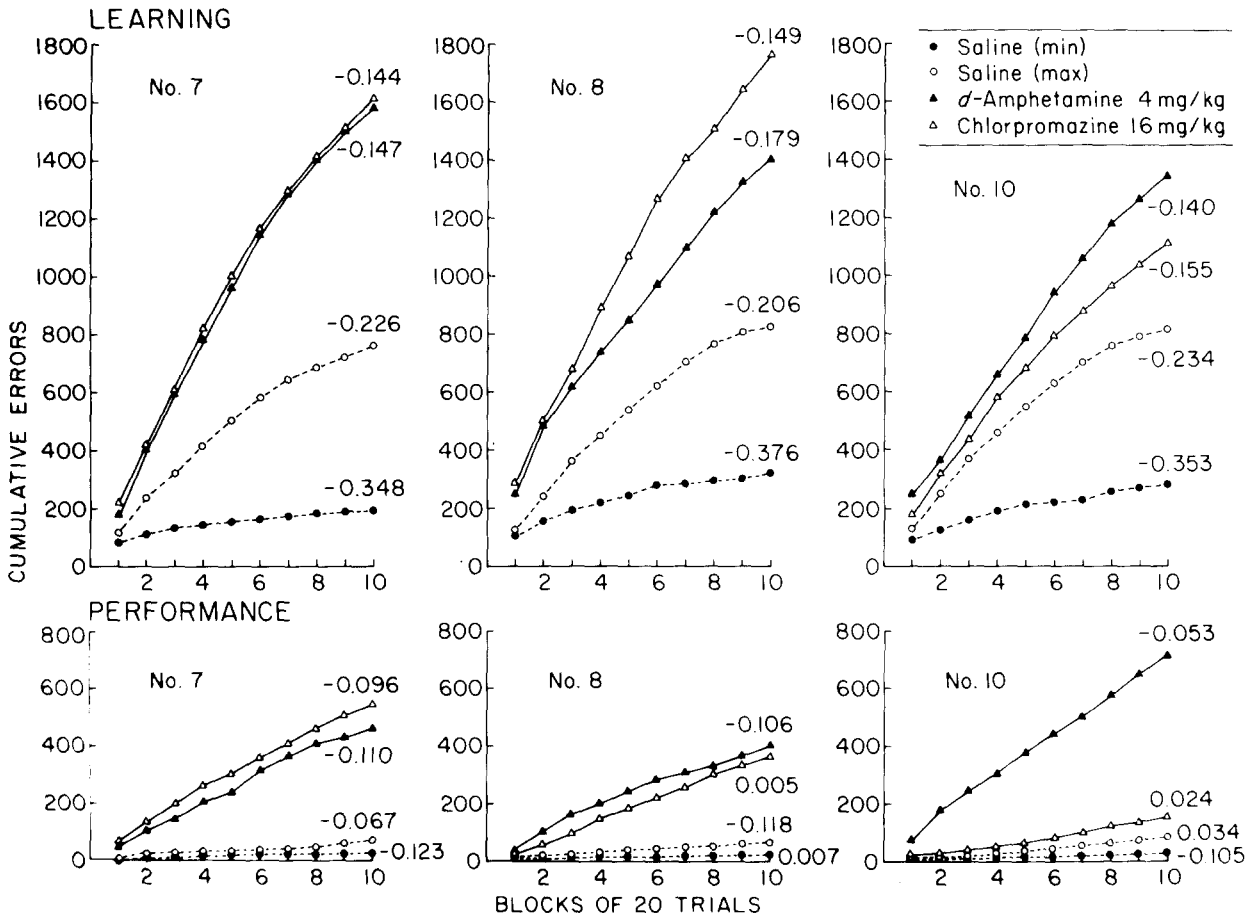

FIG. 2. Within-session effects on accuracy obtained with the largest doses of *d*-amphetamine and chlorpromazine (first determinations) under the tandem-learning and tandem-performance conditions. The saline (max) and saline (min) sessions were the sessions with the maximum and minimum total errors of all the saline sessions (16) conducted during drug testing under a given condition. The Index of Curvature value is shown for each session. The Index can range from −0.900 (maximum negative acceleration of error rate) to 0.900 (maximum positive acceleration) since each session was divided into tenths; a constant error rate yields a value of zero.

lights were off and a response had no effect. Figure 3 shows the drug effects on timeout responses for the sessions shown in Fig. 1. A comparison of Fig. 3 with Fig. 1 indicates that the effects of chlorpromazine on timeout responses were generally similar to its effects on total errors under the learning condition. The only exception occurred at the 8 mg/kg dose with No. 8, where there was an increase in timeout responses but no effect on total errors. However, under the performance condition, similar trends were obtained in only one pigeon (No. 7). With Nos. 8 and 10, there was no effect on timeout responses at any dose of chlorpromazine even though total errors increased at the higher doses. In contrast, *d*-amphetamine generally affected timeout responses and total errors differently under both the learning and performance conditions. Although total errors increased at the higher doses, timeout responses either showed no change or a slight decrease (No. 8 at 2 and 4 mg/kg under the learning condition). The only similarity between the 2 measures was found with No. 7 at the 4 mg/kg dose under the performance condition, where timeout responses were elevated by *d*-amphetamine.

DISCUSSION

The present results indicate that the repeated acquisition of tandem response sequences is affected by *d*-amphetamine in a manner similar to that found with chain sequences in previous research [25]. With both tandem and chain sequences, the largest dose (4 mg/kg) produced increases in total errors and total trial time but had little or no effect on timeout responses. In both cases, the rate of within-session error reduction (learning) was less in the drug sessions than in the control sessions. An attempt to mimic these drug effects by a prefeeding manipulation was unsuccessful [25], thereby suggesting that *d*-amphetamine's effect on learning is not related to the possible anorexic effect of the drug (the amount of grain consumed was not measured).

Despite the above similarities, however, the tandem-learning baseline appeared to be less sensitive to the effects of *d*-amphetamine than the chain-learning baseline. Doses smaller than 4 mg/kg did not increase total errors (Fig. 1) or total trial time under the tandem-learning condition but did have such effects under the chain-learning condition [25].

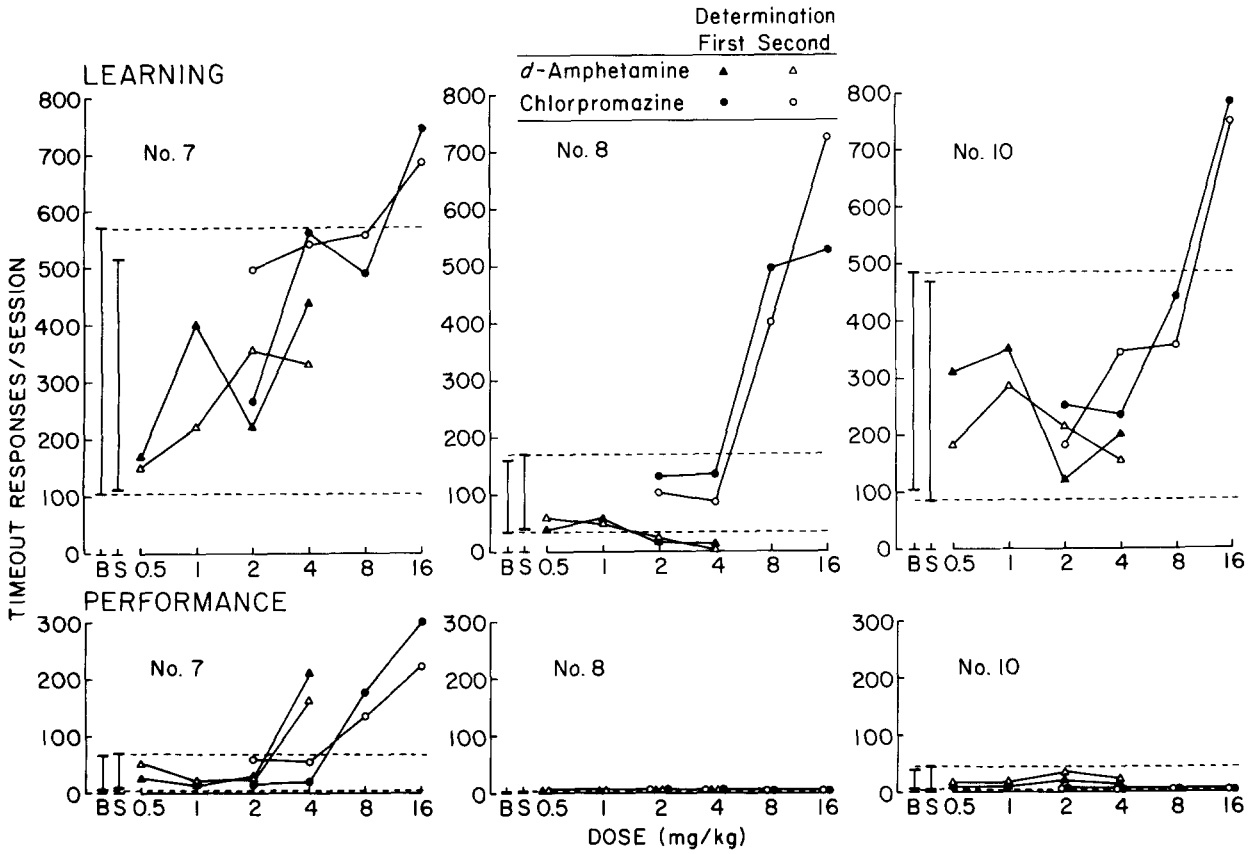

FIG. 3. Effects of *d*-amphetamine and chlorpromazine on the timeout responses per session (i.e., the total number of responses made during the 5 sec timeout periods when the lights were off and a response had no effect) under the tandem-learning and tandem-performance conditions. (See legend for Fig. 1).

The largest dose of chlorpromazine tested under the chain-learning condition was 8 mg/kg. Although this dose did not increase total errors, the within-session data suggested that ". . . if even larger doses of chlorpromazine had been tested, the overall accuracy would have been impaired" ([25], p. 512). This suggestion is supported by the present finding that 16 mg/kg of chlorpromazine produced a substantial increase in total errors (Fig. 1). The tandem-learning baseline was, however, less sensitive to the effects of chlorpromazine on total trial time than the chain-learning baseline; e.g., the 8 mg/kg dose did not increase total trial time under the tandem-learning condition but did have such an effect under the chain-learning condition [25].

Similar results have been obtained in a related "performance" situation, where two phenothiazines (chlorpromazine and trifluoperazine) had greater effects on the response rate of pigeons under a chained fixed ratio schedule than under a tandem fixed ratio schedule [24]. In a variety of other performance situations, however, it has been found that behavior under the control of external discriminative stimuli is less readily disrupted by drugs than behavior not under such control [5, 10, 11, 15, 16, 17, 19, 20, 22, 27, 28]. In discussing this apparent discrepancy, Laties [15] pointed out that ". . . framing an explanation of how drug action is modified by stimulus control may require one to determine just what types of behavioral changes are produced by the addition of particular environmental stimuli at particular times" (p. 12).

One type of behavioral change produced by switching from the chain-learning condition in the previous study [25] to the tandem-learning condition in the present research was an increase in baseline variability. Since detection of a drug effect is obviously more difficult as the control variability increases, this factor may explain why the tandem-learning baseline was less sensitive to drug effects than the chain-learning baseline. Because the drug effects originally obtained with the chain-learning baseline [25] were replicated after the completion of the present research (unpublished observations), the lesser sensitivity of the tandem-learning baseline cannot be attributed to the experimental histories of the pigeons. Although the factor of baseline variability may also account for the finding that the tandem-learning baseline was less sensitive to some of the drug effects than the tandem-performance baseline (Fig. 1), it cannot explain the different within-session changes in behavior under the two conditions (Fig. 2). Under the learning condition, the error rate decreased across trials, but the degree of negative acceleration was less (smaller Index of Curvature values) in the drug sessions than in the control sessions. In contrast, under the performance condition, the error rate was relatively constant across trials, but was higher in the drug sessions than in the control sessions.

It is difficult to compare either the tandem-learning or chain-learning baseline with other techniques reported in the literature on drugs and learning because (1) none of the previous studies of the effects of *d*-amphetamine and chlorpromazine on learning have used pigeons as subjects and (2) most of these studies have not obtained dose-effect data and have not used accuracy as a behavioral measure (see reviews: [6, 9, 13, 14, 18, 23, 29]). The tandem-performance baseline, however, may be compared with other techniques that have been used to study the dose-effects of these drugs on performance accuracy in pigeons, such as matching to sample [1, 2, 3, 12, 21] and counting schedules [4,15]. With these techniques, it has been shown that *d*-amphetamine and chlorpromazine can impair performance accuracy in a dose-related fashion, which is consistent with the present results.

In conclusion, despite certain similarities in the data obtained with chain and tandem sequences, a comparison of the present results with previous research [25] indicates that the repeated acquisition of behavioral chains is a more stable and a more sensitive baseline for assessing the effects of drugs on learning in individual animals.

REFERENCES

1. Berryman, R., M. E. Jarvik and J. A. Nevin. Effects of pentobarbital, lysergic acid diethylamide and chlorpromazine on matching behavior in the pigeon. *Psychopharmacologia* **3:** 60–65, 1962.
2. Blough, D. S. Effects of drugs on visually controlled behavior in pigeons. In: *Psychotropic Drugs,* edited by S. Garattini and V. Ghetti. Amsterdam: Elsevier, 1957, pp. 110–118.
3. Blough, D. S. Some effects of drugs on visual discrimination in the pigeon. *Ann. N.Y. Acad. Sci.* **66:** 733–739, 1957.
4. Branch, M. N. Behavior as a stimulus: joint effects of *d*-amphetamine and pentobarbital. *J. Pharmac. exp. Ther.* **189:** 33–41, 1974.
5. Carey, R. J. and R. P. Kritkausky. Absence of a response-rate-dependent effect of *d*-amphetamine on a DRL schedule when reinforcement is signaled. *Psychon. Sci.* **26:** 285–286, 1972.
6. Essman, W. B. Drug effects and learning and memory processes. *Adv. Pharmac. Chemother.* **9:** 241–330, 1971.
7. Ferster, C. B. and B. F. Skinner. *Schedules of Reinforcement.* New York: Appleton-Century-Crofts, 1957, p. 415.
8. Fry, W., R. T. Kelleher and L. Cook. A mathematical index of performance on fixed-interval schedules of reinforcement. *J. exp. Analysis Behav.* **3:** 193–199, 1960.
9. Gollub, L. R. and J. V. Brady. Behavioral pharmacology. *A. Rev. Pharmacol.* **5:** 235–262, 1965.
10. Heise, G. A. and N. L. Lilie. Effects of scopolamine, atropine, and *d*-amphetamine on internal and external control of responding on non-reinforced trials. *Psychopharmacologia* **18:** 38–49, 1970.
11. Holloway, F. A. and R. A. Wansley. Factors governing the vulnerability of DRL operant performance to the effects of ethanol. *Psychopharmacologia* **28:** 351–362, 1973.
12. Holz, W. C. Drug-rate dependency in pigeons' conditional discriminations. Paper presented at Southeastern Psychological Association Meeting, Miami Beach, April, 1971.
13. Jarvik, M. E. Effects of chemical and physical treatments on learning and memory. *A. Rev. Psychol.* **23:** 457–486, 1972.
14. Kumar, R., I. P. Stolerman and H. Steinberg. Psychopharmacology. *A. Rev. Psychol.* **21:** 595–628, 1970.
15. Laties, V. G. The modification of drug effects on behavior by external discriminative stimuli. *J. Pharmac. exp. Ther.* **183:** 1–13, 1972.
16. Laties, V. G. and B. Weiss. Influence of drugs on behavior controlled by internal and external stimuli. *J. Pharmac. exp. Ther.* **152:** 388–396, 1966.
17. Leander, J. D. and D. E. McMillan. Rate-dependent effects of drugs. I. Comparisons of *d*-amphetamine, pentobarbital and chlorpromazine on multiple and mixed schedules. *J. Pharmac. exp. Ther.* **188:** 726–739, 1974.
18. McGaugh, J. L. and L. F. Petrinovich. Effects of drugs on learning and memory. *Int. Rev. Neurobiol.* **8:** 139–196, 1965.
19. McKearney, J. W. Rate-dependent effects of drugs: modification by discriminative stimuli of the effects of amobarbital on schedule-controlled behavior. *J. exp. Analysis Behav.* **14:** 167–175, 1970.
20. McMillan, D. E. The effects of ethyl alcohol on temporally spaced responding in humans. *J. Pharmac. exp. Ther.* **171:** 159–165, 1970.
21. Mintz, D. E., D. J. Mourer and M. D. Stein. Drug effects in fixed ratio matching to sample. *Psychon. Sci.* **12:** 171–172, 1968.
22. Paasonen, M. K. and P. B. Dews. Effects of raunescine and isoraunescine on behavior and on 5-hydroxytryptamine and noradrenaline contents of the brain. *Br. J. Pharmac.* **13:** 84–88, 1958.
23. Taber, R. I. Agents affecting learning and retention of conditioned behavior. In: *An Introduction to Psychopharmacology,* edited by R. H. Rech and K. E. Moore. New York: Raven Press, 1971, pp. 213–236.
24. Thomas, J. R. Differential effects of two phenothiazines on chain and tandem performance. *J. Pharmac. exp. Ther.* **152:** 354–361, 1966.
25. Thompson, D. M. Repeated acquisition as a behavioral baseline for studying drug effects. *J. Pharmac. exp. Ther.* **184:** 506–514, 1973.
26. Thompson, D. M. Repeated acquisition of behavioral chains under chronic drug conditions. *J. Pharmac. exp. Ther.* **188:** 700–713, 1974.
27. Thompson, D. M. and P. B. Corr. Behavioral parameters of drug action: signalled and response-independent reinforcement. *J. exp. Analysis Behav.* **21:** 151–158, 1974.
28. Wagman, W. D. and G. C. Maxey. The effects of scopolamine hydrobromide and methylscopolamine hydrobromide upon the discrimination of interoceptive and exteroceptive stimuli. *Psychopharmacologia* **15:** 280–288, 1969.
29. Weiss, B. and V. G. Laties. Behavioral pharmacology and toxicology. *A. Rev. Pharmacol.* **9:** 297–326, 1969.