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Operant Methodology in the Study of Learning

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A series of experiments is described in which operant methodology is used to study the effects of drugs on "learning." Emphasis is placed on the technique of repeated acquisition as a behavioral baseline for studying this type of transition state. In this technique, each subject is required to learn a new discrimination each session. Multiple-schedule procedures are also described in which acquisition is compared to a "performance" task, where the discrimination is the same each session. The learning baseline is more sensitive to the disruptive effects of a variety of drugs (e.g., cocaine, *d*-amphetamine, haloperidol) than is the performance baseline. This general finding obtains across procedural variations and species (pigeons and monkeys). The potential usefulness of these procedures for studying both acute and chronic behavioral toxicity is discussed.

Learning has traditionally been defined as the acquisition of new behavior. Within an operant framework, however, the word learning implies transitional behavior that is progressing towards a steady state (*1*). Such transition states may reflect a variety of circumstances and behaviors. For example, a transition state may simply describe the initial acquisition of a lever-press response. Another type of transition state is the change in steady-state behavior that occurs when a single schedule of reinforcement is changed, e.g., from fixed ratio to fixed interval (*2*). A transition state may also describe the acquisition of more complex sequences of behavior, which is the focus of the present paper.

The development of an operant technique for the study of variables affecting transition states began with the work of Boren (*3*). This technique, termed repeated acquisition of behavioral chains, requires a subject to respond in a predetermined sequence on some number of operanda with a reinforcer delivered at the end of the sequence. Each session the subject is required to learn a different sequence of responses. Over time, both the pattern of acquisition and the number of errors reach a steady state from session to session. This steady state of transition states can then serve as a baseline for evaluating the effects of different independent variables by using an "individual-subject design" (each subject serves as its own control).

One type of independent variable that has frequently been studied with repeated-acquisition procedures is the administration of drugs (*4–9*). In one such procedure (*4*), a pigeon worked for food in a chamber containing three response keys, each illuminated at the same time by one of four colors. During each session the pigeon's task was to acquire a different four-response sequence (e.g., left-right-center-right) by responding on a single key in the presence of each color (see Fig. 1). An error (e.g., a response on the center key when the left key is correct) resulted in a brief timeout, during which the chamber was dark and responses had no consequences. An error did not reset the sequence; i.e., the keylights after the timeout were the same color as before the timeout.

Figure 2 shows the development of a steady state of repeated acquisition for a pigeon during baseline

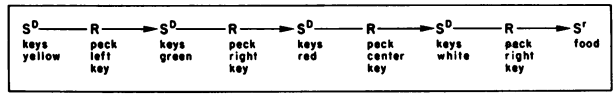

FIGURE 1. Four-response chain in a repeated-acquisition procedure.

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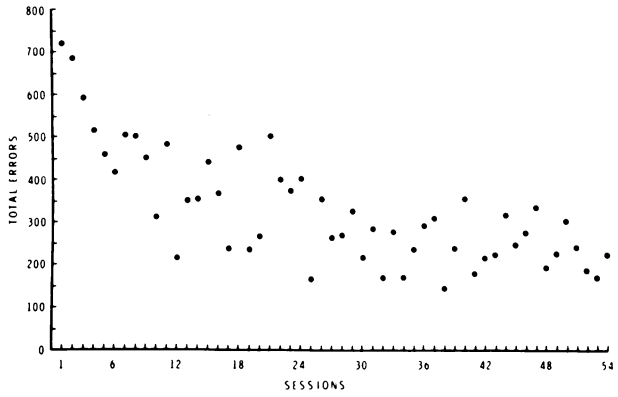

FIGURE 2. Total errors per session for a pigeon during repeated-acquisition baseline training. Data of Thompson (*10*).

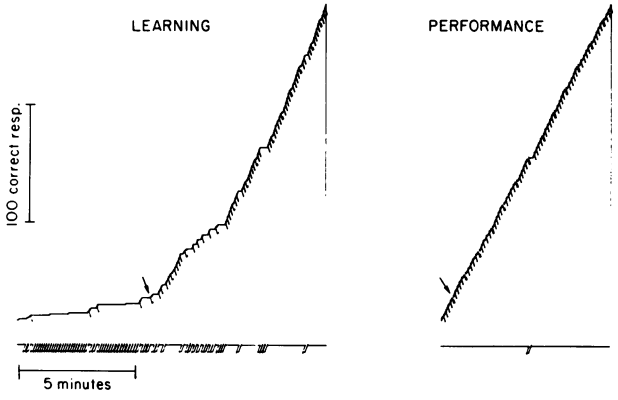

FIGURE 3. Cumulative records of a pigeon's responding under learning and performance conditions.

training (*10*). As can be seen, there was a downward trend in error levels as training continued, with the sharpest drop occurring during the first few sessions (the number of correct responses per session was held constant). The variability in error levels also decreased across sessions until a minimum range of variability was obtained (steady state). During the last 30 baseline days (sessions 25-54), almost all of the data points fell within the range of 150-350 errors. This range of variability was subsequently maintained for this subject as long as the baseline conditions were in effect.

That "learning" was produced by the repeated-acquisition procedure is illustrated in Figure 3 (left side), which shows the first part of a session for one pigeon at steady state. The response pen (top) stepped upward with each correct response and was deflected downward when the food magazine was presented (5-sec presentation for every fifth completion of the four-response chain; 0.5-sec presentation for all other completions). The arrow indicates the first food reinforcement in the session. Errors are indicated by the event pen (bottom), which was held down during each timeout. Note that errors decreased in frequency and the rate of completions of the response chain increased as the session progressed. In other words, there was an improvement of performance as a function of reinforced practice, which is an accepted behavioral definition of learning (*11*). Note also that toward the end of the record, there is an instance of nearly 80 consecutive correct responses. Since three keys were involved, the "chance" probability of this happening would be about $(\frac{1}{3})^{80}$. Under baseline conditions at steady state, essentially the same curve was obtained with each new chain this pigeon learned.

For comparison, we have also studied a performance condition, in which the four-response chain was the same from session to session. In contrast to the learning condition, the performance condition generated an error rate that was relatively constant (near zero) during the session. This is illustrated in Figure 3 (right side), which shows the first part of a performance session at steady state (the two records are from the same pigeon during different blocks of sessions).

The long-term stability of the repeated-acquisition baseline (chain-learning) is illustrated in Figure 4. For comparison, the chain-performance condition and the corresponding "tandem" conditions were also studied (*6*). Under the tandem conditions, different colored keylights were not associated with the four-response sequence; when the keylights were on, they were always white. The sessions during periods of drug testing and other experiments are omitted as indicated. Sessions 19 to 0 are the last 20 sessions under the chain-learning condition. When this condition was reinstated about a year later (Sessions 351 to 630), baseline recovery was obtained.

Some of the drug effects obtained with this repeated-acquisition procedure are shown in Figure 5 (left side), which compares the dose-effects of

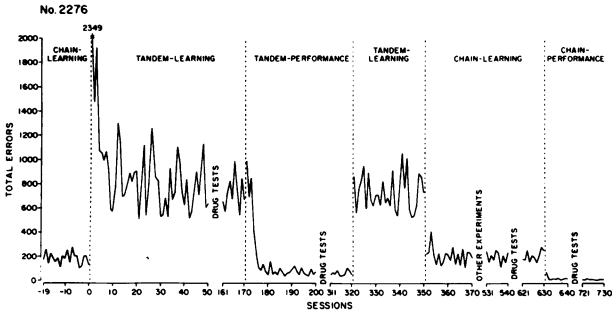

FIGURE 4. Total errors per session for a pigeon under four baseline conditions. Data of Thompson (*6*).

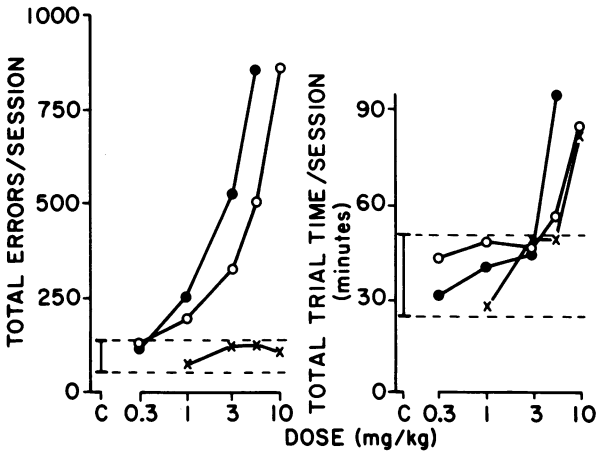

FIGURE 5. Dose effects of (●) *d*-amphetamine, (○) cocaine, and (x) fenfluramine on total errors and total trial time for a pigeon under a learning condition.

d-amphetamine, cocaine and fenfluramine (administered intramuscularly 5 min presession) on total errors per session. The doses were tested in a mixed order, one dose per week. The brackets and dashed horizontal lines indicate the control (C) ranges (based on 26 saline sessions). Only the first of two determinations for each dose is shown; the second determinations yielded similar results. Both cocaine and *d*-amphetamine increased errors as a function of dose, the only difference being that cocaine was somewhat less potent on a mg/kg basis.

In contrast, fenfluramine, which is structurally similar to *d*-amphetamine and used clinically as an "appetite suppressant," had no effect on accuracy at any of the doses tested. That fenfluramine was tested within an effective dose range is shown by its effect on total trial time (Fig. 5, right side). Total trial time (i.e., the total number of minutes that the keylights were on during a session) indicates the amount of pausing that occurred. At the highest doses, the pause-increasing effect of fenfluramine was similar to that of cocaine but less than that of *d*-amphetamine. It can also be noted that the error-increasing effect of *d*-amphetamine and cocaine occurred at doses (1 and 3 mg/kg) that had no effect on pausing. The finding that accuracy was impaired by cocaine and *d*-amphetamine but not by fenfluramine was also obtained under the chain-performance condition, although higher doses were required to detect the effects (not shown). It is interesting that this finding complements the results obtained in self-administration research. It is well established that cocaine and *d*-amphetamine can serve as reinforcers to maintain self-administration behavior in monkeys, whereas fenfluramine is ineffective in this animal model of drug abuse (*12*).

The steady state of repeated acquisition also provides a convenient means for assessing the effects of chronic administration of drugs on learning. For example, Figure 6 (top) shows some data obtained during a 50-day period in which cocaine was administered (intramuscularly 5 min presession) to pi-

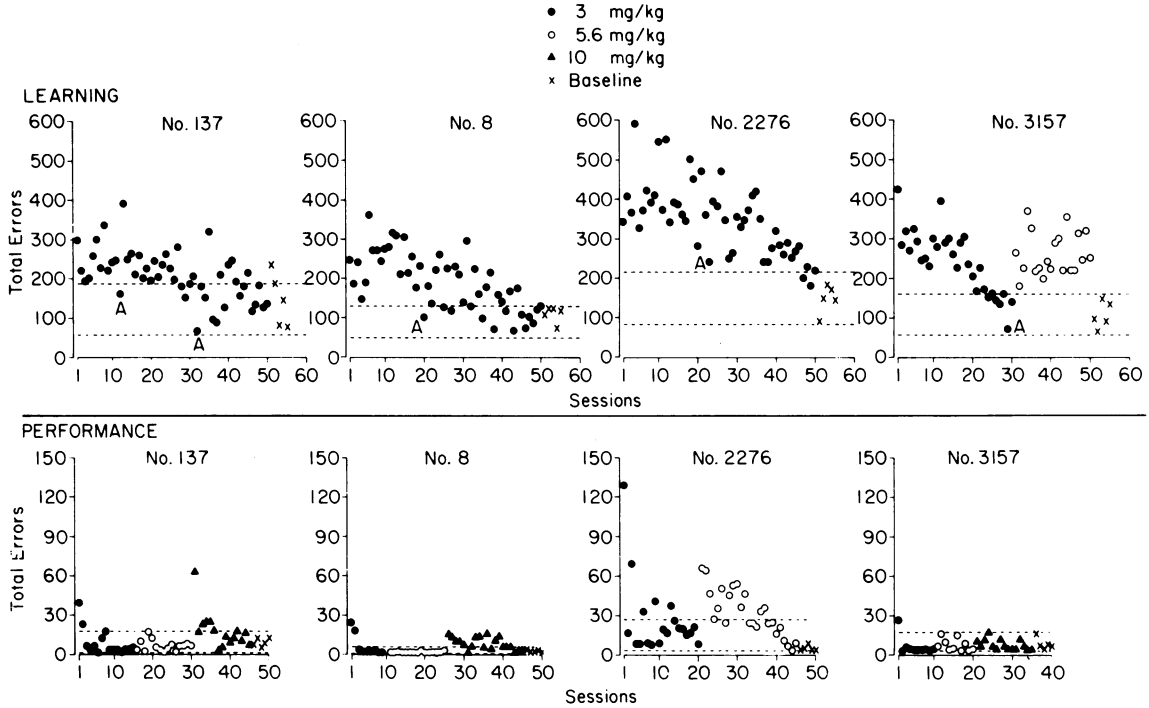

FIGURE 6. Effects of chronically administered cocaine (3, 5.6 or 10 mg/kg/day) on total errors per session for four pigeons under learning and performance conditions. Data of Thompson (*8*).

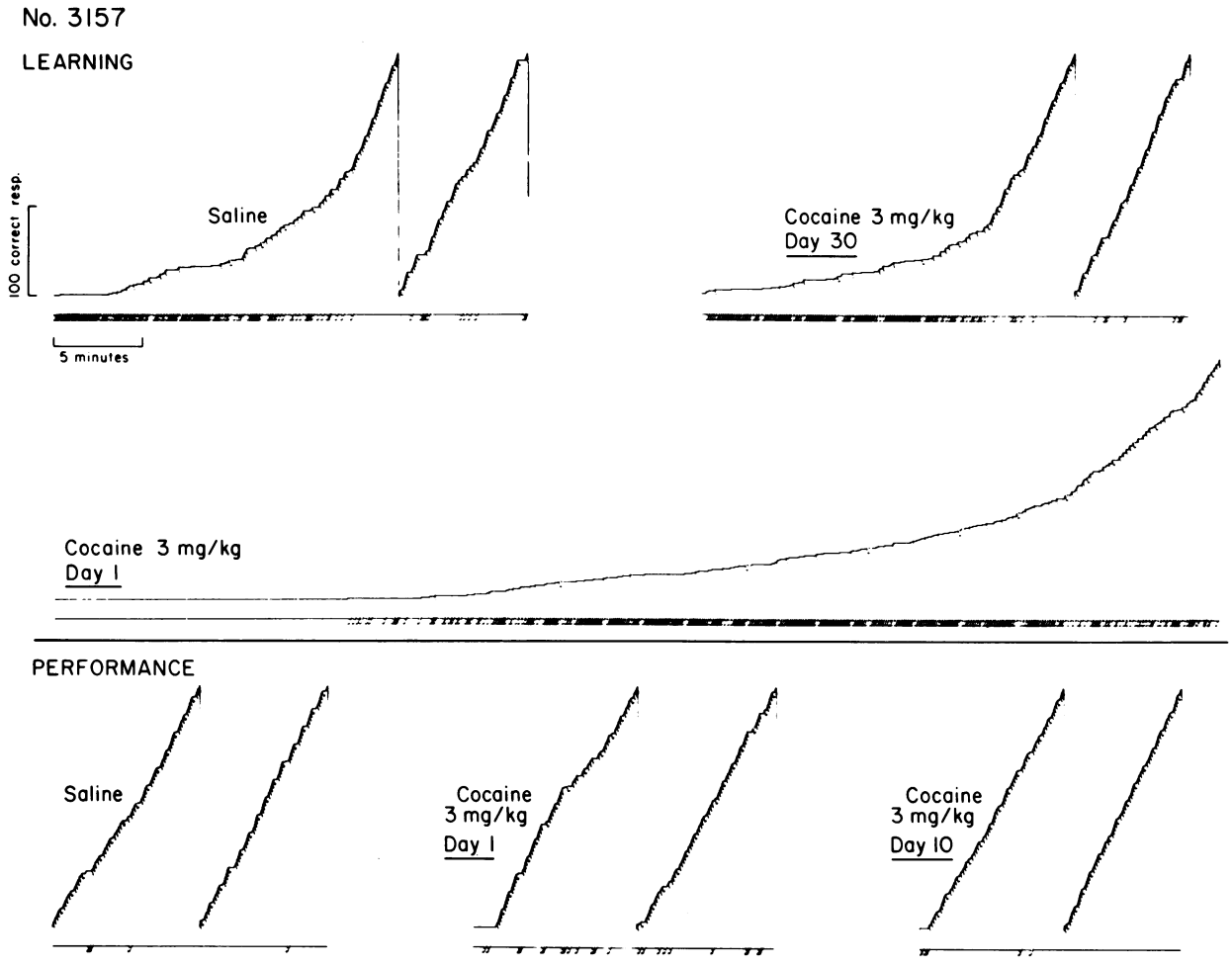

FIGURE 7. Cumulative records showing the development of behavioral tolerance to cocaine (3 mg/kg/day) for a pigeon under learning and performance conditions. Data of Thompson (*8*).

geons (*8*). The two dashed horizontal lines for each pigeon indicate the control range of variability (based on 10 saline sessions that preceded the chronic drug regimen). The data points marked A indicate the sessions in which a chain was repeated after only one intervening session involving a different chain. Under the learning condition, the initial administration of 3 mg/kg of cocaine increased the total errors per session in all four pigeons. The error-increasing effect did not persist, however, during the 30 to 50 sessions of repeated administration of this dose; the total errors tended to decrease across sessions until they fell within the control range, i.e., behavioral tolerance developed. When the dose was increased to 5.6 mg/kg for no. 3157, the total errors increased and stayed above the control range for the remainder of the chronic drug regimen (20 sessions). The lack of tolerance at 5.6 mg/kg is in contrast to the downward trend in the data points of no. 3157 during the first 20 sessions at 3 mg/kg. In general, when the chronic cocaine regimens were discontinued, the behavior returned to or remained within the control ranges (see baseline data).

For comparison, cocaine was also administered chronically under the performance condition, where the chain of correct responses was the same from session to session. Under the performance condition (Figure 6, bottom), the initial administration of 3 mg/kg of cocaine increased errors in all four pigeons as in the learning condition. However, with repeated administration of this dose, tolerance developed very quickly, i.e., after 1 to 3 sessions, performance errors returned to within the control range and generally remained there. In other words, accuracy under the performance condition was less readily disrupted by chronically administered cocaine than accuracy under the learning condition.

This was true regardless of whether the drug was tested first under the learning condition (nos. 137 and 2276) or under the performance condition (nos. 8 and 3157). Increasing the dose of cocaine to 5.6 mg/kg had no effect on performance errors in three of the pigeons. The absence of a disruptive effect of 5.6 mg/kg in no. 3157 under the performance condition is in striking contrast to the error-increasing effect of this dose under the learning condition. With no. 2276, 5.6 mg/kg of cocaine initially produced an increase in performance errors but tolerance developed during repeated administration of this dose (25 sessions). Note that tolerance developed more slowly at 5.6 mg/kg than at 3 mg/kg. There was also an initial error-increasing effect and the development of tolerance in two of the pigeons (nos. 137 and 8) when the dose was further increased to 10 mg/kg.

Figure 7 shows some within-session effects of cocaine (*8*). The cumulative records are from a representative saline session of no. 3157 and the first and last drug sessions at 3 mg/kg under the learning and performance conditions. The first two excursions of the response pen in each session are shown, except for day 1 under the learning condition, where only the first excursion is shown. The recording details are the same as in Figure 3. Under the learning condition (saline), the errors decreased in frequency and the rate of completions of the response chain increased as the session progressed. During the first cocaine session, there was an initial period of pausing and then a long period in which errors occurred at a high frequency. It is evident that tolerance to the drug-induced increase in errors and pausing had developed by the last session (day 30). In contrast to the learning condition, the performance condition (saline) generated an error rate that was relatively constant (near zero) during the session. Cocaine (day 1) initially produced a brief pause followed by an increased error rate. The record from day 10 shows that tolerance developed to these effects of cocaine. That tolerance developed more quickly under the performance condition than under the learning condition is consistent with the widely held view that behavior under strong stimulus control is less readily disrupted by a drug than behavior under weak stimulus control (*13, 14*). That the baseline error levels under the performance condition were much lower than those under the learning condition indicates that the control by the discriminative stimuli (e.g., keylight colors, timeout) was stronger under the performance condition, where the response sequence was the same

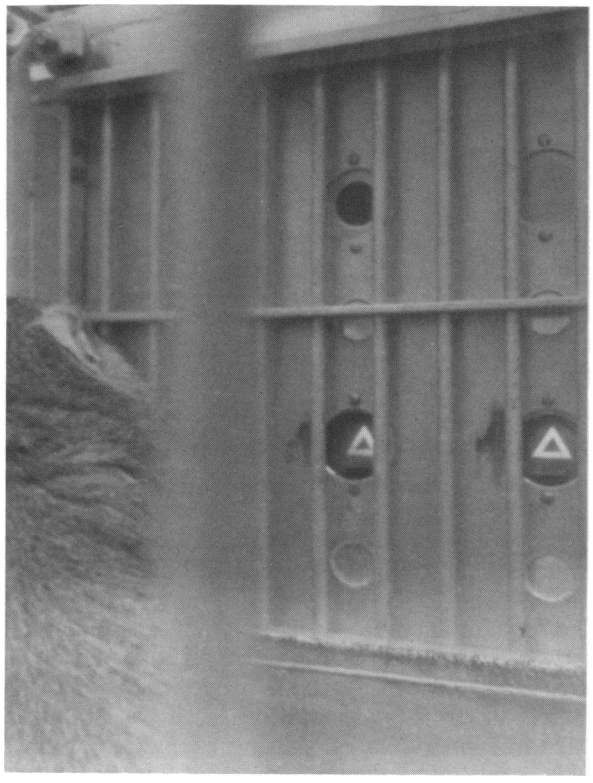

FIGURE 9. Interior view of primate cage with response panel in place.

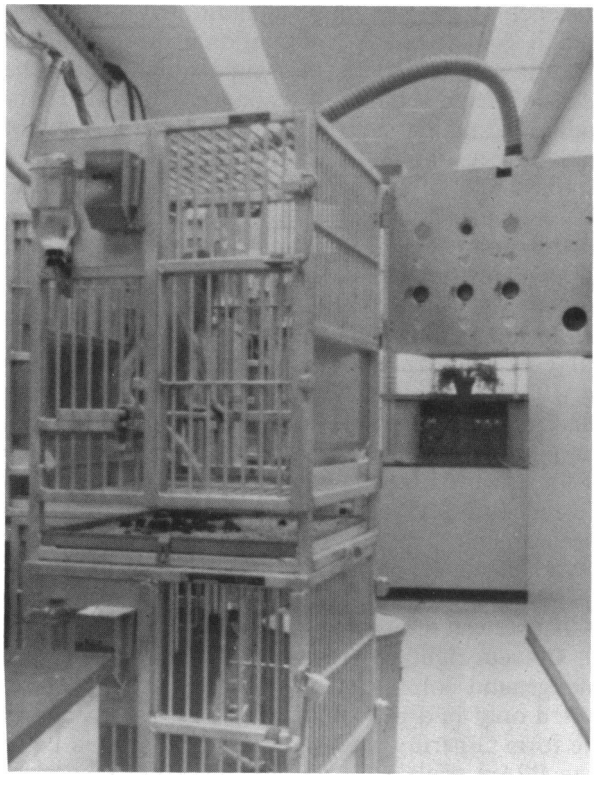

FIGURE 8. Primate cage with removable response panel.

from session to session.

A methodological means by which differential drug effects on learning and performance can be evaluated during the same session is through the use of a multiple schedule of reinforcement. A multiple schedule is defined as "a compound schedule in which two or more component schedules operate in alternation, each in the presence of a different stimulus" (*15a*). In a multiple schedule we are using with monkeys, one component is a repeated-acquisition baseline and the other component is a performance condition. Each monkey *(Erythrocebus patas)* is individually housed in a standard primate cage (Lab-Care Caging). A removable response panel (BRS/LVE TIP-001) is attached to the side of each subject's cage during the experimental session (see Figs. 8 and 9). Mounted on this panel is a row of three press plates, behind which are mounted stimulus projectors and a pellet feeder. The subject can easily be restrained (via a squeeze mechanism) for injection.

The repeated-acquisition component is similar to that described previously for the pigeons. One of four geometric forms (horizontal line, triangle, vertical line, circle) is projected on a red background on all three press plates. During each session the monkey's task is to acquire a different four-response sequence by responding on a single press plate in the presence of each form. The other component of the multiple schedule is a performance condition, where the four-response sequence remains the same from session to session. During the performance component, the four geometric forms are projected on a green background. In both learning and performance, the completion of every fifth sequence is reinforced with food (FR 5); each error produces a brief timeout, during which all stimuli are turned off. The two components alternate after 10 reinforcements or 15 min, whichever occurs first.

The type of baseline behavior generated by the multiple-schedule procedure is illustrated in the top cumulative record of Figure 10. Each correct response stepped the pen upward and each completion of the four-response sequence deflected this pen. Errors are indicated by the lower event pen. A change in components of the multiple schedule reset the stepping pen. As can be seen, during the first learning component (L), errors decreased in frequency and the rate of completions of the sequence increased. Following the tenth reinforcement in the learning component, the pen reset, and the performance component (P) began. Note that no errors were made in the performance component, an indication of strong stimulus control. Following the tenth reinforcement in the performance component, the schedule changed back to the learning component. This alternation continued throughout the session. By the end of the session, the behavior in the two components was virtually identical.

The effects of 0.56 mg/kg of cocaine (administered intramuscularly 5 min presession) are shown in the second cumulative record of Figure 10. As can be seen, the monkey did not respond at all during the first learning component (weak stimulus control). However, as soon as the performance component began (strong stimulus control), correct responding occurred at a high rate without any errors being made. A similar pattern of pausing and high-rate responding occurred during the second learning and performance components, respectively. As the session progressed, the rate of correct responding increased across successive learning components, but a selective error-increasing effect was also evident; accuracy in performance remained unaffected. The selective drug effects obtained with the multiple schedule are similar to those obtained when repeated acquisition and performance were studied separately (Fig. 7). In both cases, cocaine had greater disruptive effects (increased errors and pausing) on learning than on performance.

Another procedure we have used involves the acquisition of conditional discriminations. A discrimination where the reinforcement of a response in the presence of one stimulus is conditional upon other stimuli is defined as a conditional discrimination (*15*). In such situations, no single stimulus sets the occasion for a reinforced response. The basic procedure is summarized in Figure 11. At the start of each trial, a stimulus (e.g., cross-red) was displayed on the center key. A peck on the center key (i.e., an R1 response) illuminated the two side keys white. At this point the pigeon's task was to peck one of the two side keys, depending upon the stimulus displayed on the center key. A response to either side key terminated the trial and turned the side keys off. All correct responses resulted probabilistically ($p = 0.75$) in a change of the geometric form superimposed upon the background color. That is, following a correct response the occurrence of each of four forms was equiprobable on any given trial. Correct left-key responses advanced the chain to the next link. The first correct response on the left key changed the background color from red to yellow (see R3 on trial 2). The second correct left-key response was reinforced with grain (see R3 on trial 5). Correct right-key responses did not change the background color. Correct right-key responses resulted only in a probabilistic ($p = 0.75$) change of the form superimposed upon the background color (see R2 on trials 1, 3, and 4). Incorrect responses made on either the left or right key resulted in a brief timeout, during which the chamber was dark

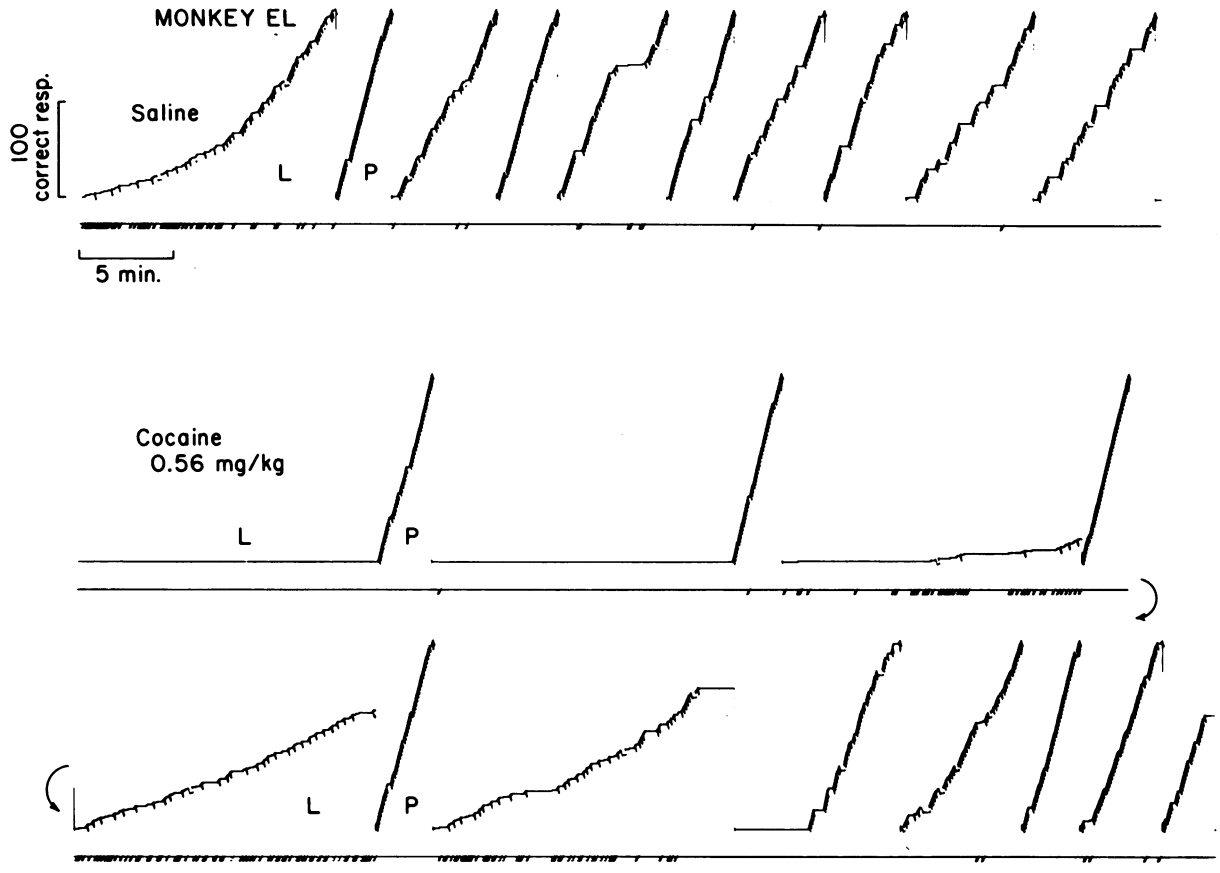

FIGURE 10. Effects of cocaine (0.56 mg/kg) on a monkey's responding on a multiple schedule with learning (L) and performance (P) components.

(e.g., R3 on trial 1 and R2 on trial 2). Following a timeout the same stimulus was presented on the subsequent trial(s) until a correct response was made (correction procedure).

In summary, in this conditional discrimination procedure, the subject was required to respond to different combinations of colors and forms, responding on the left key in the presence of two different discriminative stimuli and on the right key in the presence of any of six different stimuli. The requirements for food reinforcement were the identification (i.e., left-key response) of two discriminative stimuli and the rejection (i.e., right-key response) of a variable number of discriminative stimuli.

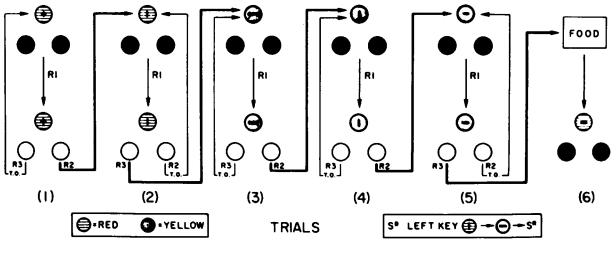

FIGURE 11. Conditional discrimination procedure.

This procedure constituted the basic behavioral task in each of two components of a multiple schedule. In one component, the chain of conditional discriminations remained the same from session to session (performance). In the other component, however, the chain of conditional discriminations changed from session to session (learning). Specifically, the form associated with each color, which set the occasion for a left-key response, changed with each session. A more detailed description of this conditional-discrimination procedure may be found elsewhere (*16*).

The effects of cocaine on the behavior of a pigeon responding on this schedule are shown in Figure 12. In these records, correct responses stepped the pen upward and reinforcement is indicated by a downward deflection of the same pen. Downward deflections of the lower event pen indicate timeouts (i.e., errors). A change in components of the multiple schedule reset the stepping pen, while the solid de-

flections of the lower pen indicate a brief delay that separated the component changes. A representative record for a saline session is shown at the top of Figure 12. The session began with the learning component (L). As is shown in the cumulative record, the frequency of errors rapidly decreased within this initial learning component. This abrupt pattern of acquisition was typical of this subject. Following the tenth reinforcement in the learning component, the first performance component (P) began. Note that no errors were made during this component. Following the tenth reinforcement, the schedule changed back to the learning component. This alternation continued until a total of 60 reinforcements were delivered (three components each of learning and performance). As can be seen in the record, no errors were made in performance, while errors decreased across the three learning components. In comparison to this control record, the effects of cocaine (4.2 mg/kg, administered intramuscularly 5 min presession) are shown in the middle of Figure 12. As is evident from the cumulative record (day 1), considerable periods of time elapsed during which no responding occurred. When responding did occur in the learning components, however,

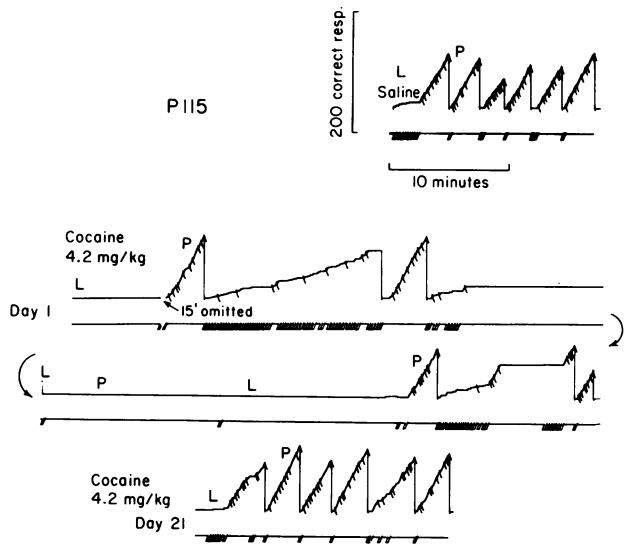

FIGURE 12. Effects of chronically administered cocaine (4.2 mg/kg, day 1 and day 21) on a pigeon's responding on a multiple schedule with learning (L) and performance (P) components.

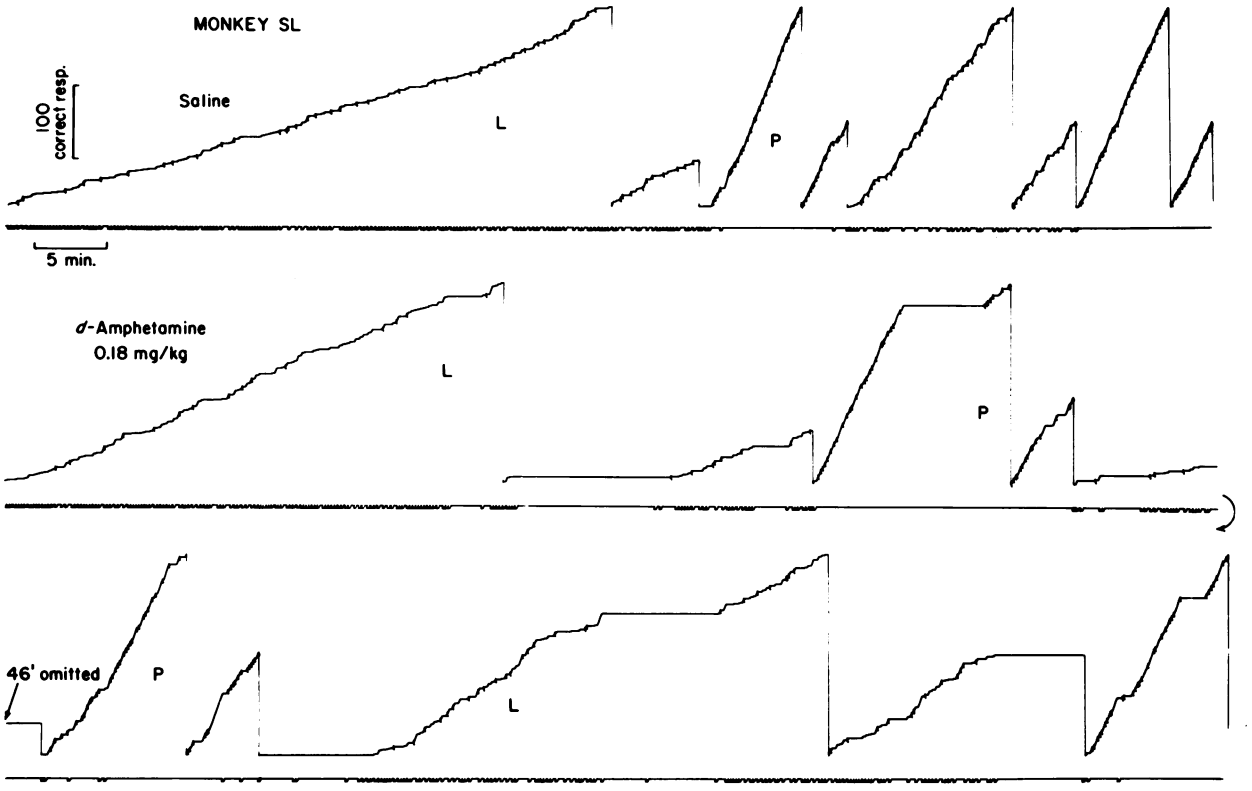

FIGURE 13. Effects of *d*-amphetamine (0.18 mg/kg) on a monkey's responding on a multiple schedule with learning (L) and performance (P) components.

cocaine increased errors in comparison to control. Furthermore, the normal pattern of within-session acquisition observed under control conditions was virtually eliminated by cocaine. In sharp contrast to this error-increasing effect in learning, errors did not increase in the performance components. This selective drug effect on the learning and performance of conditional discriminations is similar to that obtained with the repeated-acquisition procedure previously described (see Fig. 10). The effects of cocaine after 21 days of repeated administration are shown in the bottom cumulative record (day 21) of Figure 12. Note that in comparison to day 1, both the pausing and error-increasing effects of the drug have largely disappeared, thus indicating the development of behavioral tolerance to cocaine.

The cumulative records in Figure 13 illustrate the effects of *d*-amphetamine (0.18 mg/kg, administered intramuscularly 15 min before the session) on a monkey's responding on a similar multiple schedule (learning and performance) of conditional discriminations. In this experiment, a correction procedure was not used and the number of possible conditional discriminations was increased. Note that the behavior during the control session (top) is qualitatively similar to that seen in Figure 12; i.e., few errors occur in the performance components while errors decrease within the session in the learning components. As can be seen in the lower half of Figure 13, a selective drug effect on learning is again evident. *d*-Amphetamine increased errors in the learning component while having virtually no effect on accuracy in performance. Drug-induced pauses in responding also were more frequent and prolonged in the learning than in the performance component.

Another operant procedure for studying acquisition involves "fading the stimulus" (*15b*). This procedure has been used for eliminating the control by a stimulus without disrupting behavior (*17*). In order to investigate the effects of drugs on this type of acquisition, we have incorporated a fading procedure as a third component of a multiple schedule of learning and performance. Similar to the procedures described earlier, a different four-response sequence constituted the behavioral task in each component. In each component, the subject was required to emit a sequence of four responses, in a predetermined order, on four levers with food reinforcement delivered at the end of the sequence. Errors produced a brief timeout, during which all stimuli were turned off. Different stimuli (e.g., red, blue, and green) were associated with each component of the multiple schedule. Unlike the previous procedures, however, different discriminative stimuli were not associated with each response in the sequence; i.e., a "tandem" sequence was used.

The first component of the multiple schedule was a repeated-acquisition task, where the sequence of correct responses changed from session to session. In the second component of the multiple schedule, the sequence of correct responses remained the same from session to session (performance). The third component of the multiple schedule was also a repeated-acquisition task, but acquisition in this component was supported through the use of a stimulus-fading procedure. In the first step of this fading procedure, a lamp would light (sequentially) only over the correct lever. In subsequent steps, the illumination of the lamps over the incorrect levers increased, until at the final step the lamps over all the levers were illuminated equally. Completion of a correct sequence advanced the fading level one step. Four errors within a single sequence decreased the fading level one step.

The behavior of a monkey responding on this schedule is shown in the cumulative record at the top of Figure 14 (saline). Each response stepped the pen upward and reinforcement is indicated by a downward deflection of the same pen. Downward deflections of the lower event pen indicate timeouts (i.e., errors). The event pen was also deflected (solid deflections) and the stepping pen reset when a component change occurred. The components changed after either 20 reinforcements or after 25 minutes (excluding timeouts), whichever occurred first. As is shown in the cumulative record, the session began in the learning component (L), then changed to performance (P), which was then followed by the faded learning component (FL). The individual components alternated in this order (L-P-FL) throughout the session. Notice in the control record that the greatest number of errors occurred in the learning component and the fewest (only one) in performance. Although within-session acquisition occurred in both the learning and faded learning components, acquisition was facilitated (fewer errors) by the fading procedure. The effects of haloperidol (0.0075 mg/kg, administered intramuscularly 30 min presession) are shown in the second cumulative record of Figure 14. At this dose, haloperidol selectively increased total errors in the learning component. Acquisition with the drug was incomplete in comparison to saline in both learning components. This effect can be seen in the cumulative records by comparing the frequency of errors at the end of the drug and control sessions. The effects of a higher dose (0.01 mg/kg) of haloperidol are shown in the third cumulative record of Figure 14. As can be seen in this record, the subject responded at the beginning of the session (L) and then paused approximately 115 min, during

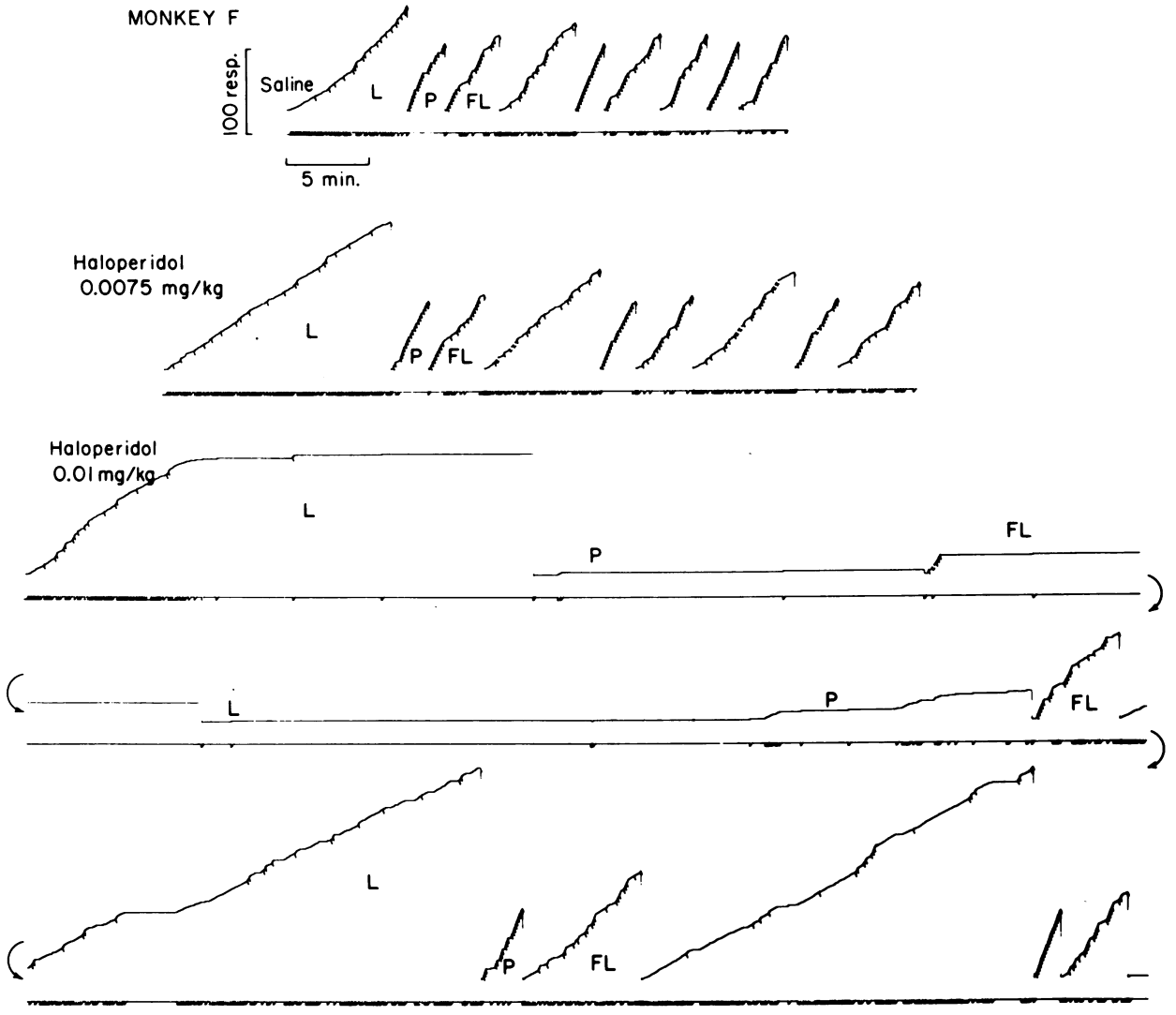

FIGURE 14. Effects of haloperidol (0.0075 mg/kg and 0.01 mg/kg) on a monkey's responding on a multiple schedule with learning (L), performance (P) and faded learning (FL) components.

which few responses were made. When the subject began responding again, errors increased in each of the three components. The largest error-increasing effect occurred in the learning component (L). As is apparent from the bottom record, acquisition was virtually eliminated in this component. Though incomplete in comparison to control, within-session acquisition occurred in the faded learning component (FL). In contrast, by the end of the session, responding in the performance component (P) was indistinguishable from that of control.

In conclusion, we have shown that repeated-acquisition procedures provide sensitive baselines for assessing the effects of drugs on learning in individual subjects. The generality of our findings was established by showing that the drug effects were similar across procedural variations and species (pigeons and monkeys). By incorporating the repeated-acquisition technique into a multiple schedule, it was possible to make a direct comparison of a drug's effect on learning and performance within the same session. In general, we have found the learning condition to be more sensitive to drug effects than the performance condition. Just as we have observed differential sensitivity to the effects of drugs on learning and performance, we would expect these baselines to be differentially sensitive to the behavioral effects of low doses of toxic substances. For example, at low concentrations, acute behavioral toxicity may be restricted to the learning component, whereas at higher concentrations, both learning and performance may be impaired. Simi-

larly, the steady states of repeated acquisition and performance are well suited for the study of chronically administered agents. For example, the initia administration of a suspected toxin may affect onl behavior in the learning component. However, witl repeated administration, a cumulative behaviora toxicity may be encountered where the agent affect both learning and performance. While this operan methodology for studying learning may not be use ful in the initial screening of drugs or toxic sub stances because of the time required to establisl baseline stability, the methodology does permit a assessment when more complex forms of behavio are of interest.

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