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Behavioral research in pigeons with ARENA: An Automated Remote Environmental Navigation Apparatus

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ABSTRACT

Three experiments established the effectiveness of an Automated Remote Environmental Navigation Apparatus (ARENA) developed in our lab to study behavioral processes in pigeons. The technology utilizes one or more wireless modules, each capable of presenting colored lights as visual stimuli to signal reward and of detecting subject peck responses. In Experiment 1, subjects were instrumentally shaped to peck at a single ARENA module following an unsuccessful autoshaping procedure. In Experiment 2, pigeons were trained with a simultaneous discrimination procedure during which two modules were illuminated different colors; pecks to one color (S+) were reinforced while pecks to the other color (S−) were not. Pigeons learned to preferentially peck the module displaying the S+. In Experiment 3, two modules were lit the same color concurrently from a set of six colors in a conditional discrimination task. For three of the colors pecks to the module in one location (e.g., upper quadrant) were reinforced while for the remaining colors pecks at the other module (e.g., lower quadrant) were reinforced. After learning this discrimination, the color-reinforced location assignments were reversed. Pigeons successfully acquired the reversal. ARENA is an automated system for open-field studies and a more ecologically valid alternative to the touchscreen.

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It is integral to science that any measure of observed behavior be both accurate and reliable. Notable advances in the reliability of behavioral measurement in the psychological and neuroscience literature include the development of the operant box connected to a cumulative recorder (e.g., Skinner, 1938, 1956), of tracking systems for spatial behavior (e.g., Clarke et al., 1985; Noldus et al., 2001), and the use of touchscreen-equipped operant boxes for pigeons (e.g., Allan, 1992; Blough, 1986; Pisacreta and Rilling, 1987; Wright et al., 1988), rats (e.g., Bussey et al., 1994; Cook et al., 2004; Markham et al., 1996; Sahgal and Steckler, 1994), and primates (e.g., Elsmore et al., 1989). Each apparatus improved reliability by using the latest technology to automate data collection. Additionally, the use of an operant box minimized the contribution of handling effects to variance in the data by presenting continuous trials. Some of these technologies, however, resulted in more contrived, less naturalistic settings. For instance, two-dimensional depictions of objects or scenes with color properties designed to suit trichromatic primates rather than pentachromatic avians likely appear less natural to a pigeon than do the real, three-dimensional objects or scenes they represent (e.g., Cabe, 1976; Cole and Honig, 1994; Friedman et al.,

2005; Zeigler and Bischof, 1993; but see also Spetch and Friedman, 2006a,b). The degraded ecological validity is often a concern for the generalization of findings collected using these technologies to a larger population or different contexts.

Open-field studies continue to be used as a more naturalistic alternative to the operant box. Open-field studies have been used to investigate behavior in a variety of species, including humans, cats, dogs, rats, and pigeons, and using dependent measures involving whole body movements, partial body movements, and adrenal and electrophysiological activity (see Walsh and Cummins, 1976 for a review). Although widely used, Walsh and Cummins concluded that the construct validity and reliability of many of these dependent measures for open-field study remain unproven and that this failure undermines the interpretations and conclusions of the studies based upon them. Even the literature reporting on a single species (e.g., pigeons) in an open field indicates that few researchers adopt identical criteria for their dependent measure; this inconsistency complicates comparisons across different laboratories. Another disadvantage to using the traditional open field is that the number of trials given and subjects tested per day is severely limited by labor demands and the time consuming nature of a discrete-trial procedure. These limitations have resulted in few open-field studies which have parametrically varied the independent variable to more completely evaluate its relationship to behavior.

We have found data collection in open-field studies in our own lab to be constrained by two factors. The first constraint is the

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placement of food rewards directly in the open-field setting. Food goals must be replaced or at least moved to new locations after each trial. Second, landmarks or other types of discriminative stimuli placed directly in the open-field setting must be relocated to new positions prior to each trial. Both of these procedures necessitate the removal of the subject from the open field, thereby introducing handling effects and disrupting the smooth progression of the subject's behavior. A number of studies have tried to address these issues through the use of a start box (e.g., Biegler and Morris, 1999; Lechelt and Spetch, 1997) or specialized reward location (e.g., Biegler and Morris, 1999; Cheng, 1988), but to our knowledge no open-field apparatus has combined automation of reward delivery and stimulus presentation to create a fully automated system.

Recently, Badelt and Blaisdell (2008) reported the development of a circuit using a capacitive sensor for detecting proximity and response of a subject. This sensor can be incorporated into a variety of configurations and adapted to numerous apparatus to automate response detection. For example, Badelt and Blaisdell tested the validity and reliability of the circuit in detecting perching behavior of pigeons, the passage of rats and mice through an alley maze or open field, and the detection of a pigeon pecking or a rat nose poking into a small cup. In the apparatus for pigeons, the circumference of the cup (9.4 cm) was a little greater than that of a standard United States of America quarter (8.0 cm) and the depth of the cup was equal to its diameter (3.0 cm). The cup was embedded in the center of a small plastic box (see Fig. 1). The plastic box housed a circuit board, sensor electrodes, a set of light-emitting diodes (LEDs), batteries, and a wireless transmitter and receiver. The LEDs protruded into the bottom of the cup and served to illuminate the area when activated by the computer. We have built a number of these boxes which can be controlled remotely by a computer to both detect subject responses and to display colored lights from the LEDs. When placed on the floor of an open field, these devices can be used to automate a variety of behavioral procedures, ranging from simple Pavlovian and operant conditioning to spatial and temporal cognition to patterning and rule learning. The entire behavioral system, called ARENA (Automated Remote Environmental Navigation Apparatus), consists of a holding cage with a food hopper, an automatic door, and the ARENA modules placed on the floor of the open field. This system has the potential to resolve many of the remaining issues discussed above that involve the collection and scoring of behavior in an open field.

Here we report a series of experiments designed to test the efficacy of this new technology. The use of this new technology within an open-field setting (1) improves the objectivity of the behavioral measure since the animal's responses are detected via the apparatus and data recording does not depend on interpretation by an observer, (2) eliminating handling of the subject in between trials minimizes handling-induced stress and agitation; for example, in pilot studies using a landmark-based task in a traditional open field subjects would frequently remain stationary for several minutes at the start of a trial before reengaging in foraging behavior, (3) stimuli can be changed instantaneously which reduces labor demands on the experimenter, and (4) automated reward delivery at a single location eliminates the influence of visual or odor cues at the site of reward (e.g., food buried by the experimenter). These benefits combine to enable many more trials to be conducted in a single session; in the past a session with 80 trials of a landmark-based search task in the open field was not feasible but can be completed in ARENA in 1 h.

We tested pigeons in three basic conditioning procedures—autoshaping/instrumental shaping, a simultaneous discrimination, and a conditional discrimination. We selected these procedures because they are among those most commonly used to study basic behavioral and cognitive processes in laboratory animals. All three of these procedures involve presentation of visual stimuli
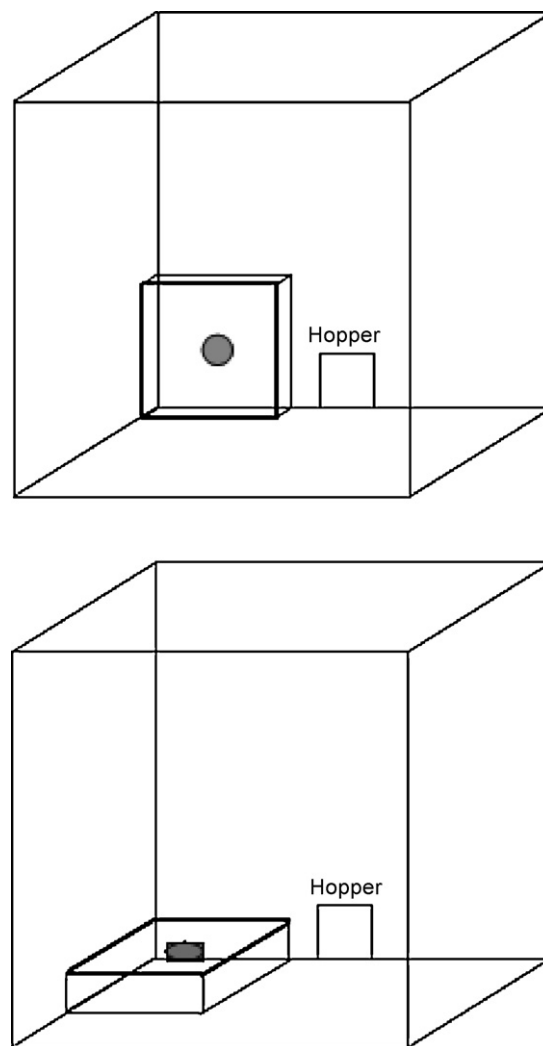

Fig. 1. A schematic representation of the operant box used in Experiment 1 and the placement of the ARENA module. The top panel shows the module vertically positioned, while the bottom panel shows the module in the horizontal position. The grey filled circle indicates the stimulus-response area. 'Hopper' indicates the location of food reinforcement. Figure not drawn to scale.

and detection of subject response, and probe the development of behavior in response to stimulus-outcome or response-outcome contingencies. Furthermore, these procedures involve the continuous presentations of trials as in more conventional studies conducted in the operant box.

1. Experiment 1

This experiment used autoshaping (a.k.a., sign tracking) and instrumental shaping procedures to determine whether subjects would interact with a single ARENA module in a manner that would make it useful for behavioral studies. An autoshaping procedure involves the development of a Pavlovian conditioned response to a signal (conditioned stimulus or CS) of a motivating stimulus (unconditioned stimulus or US) such as the delivery of food (Brown and Jenkins, 1968). Notably, the delivery of the US does not depend on the subject's response, and thus any response to the CS that develops is a true Pavlovian conditioned response. An instrumental shaping procedure is similar except that a subject's response is necessary for the delivery of the reward.

One critical component of the ARENA module to be tested was the reliability of the capacitive sensor to detect responses to the

cup. Unlike conventional methods of behavioral measurement that require physical interaction between the subject and the measuring device, a capacitive sensor is triggered by the mere proximity of the subject's free-space capacitance within the sensor's field of detection. Thus, the subject does not receive tactile feedback that it has successfully activated the sensor. Badelt and Blaisdell (2008) demonstrated the reliability of a capacitive sensor to detect subject responses in a small set of trials in a single session. The current experiment extends the reliability test across a large number of sessions. This experiment also tested the functional utility of LED illumination as a signal for the food US which was delivered from a nearby hopper. Each trial consisted of a brief presentation of white light from the ARENA module followed immediately by the delivery of grain and pellets reinforcement. Across sessions, we manipulated the position of the module, the duration and intensity of the light cue, the reward schedule, and the Pavlovian or operant nature of the contingency.

1.1. Method

1.1.1. Subjects

Five white Carneaux pigeons (*Columba livia*; Double T Farm, IA) served as subjects. Subjects had previously served in an autoshaping and in a spatial-search procedure in a touchscreen box. Pigeons were maintained at 80–85% of their free-feeding weights. They were individually housed in a colony with a 12-h light–dark cycle and had free access to water and grit. Experimental procedures occurred during the light portion of the cycle.

1.1.2. Apparatus

Testing was conducted in a flat-black Plexiglas chamber (38 cm wide × 36 cm deep × 38 cm high) illuminated at all times by a 28 V, 2.2-W bulb located in the ceiling. A food hopper (Coulbourn Instruments, Allentown, PA) was located in the center of the front panel, its access hole flush with the chamber floor. When the hopper was activated to the raised position a 28 V, 2.2-W bulb illuminated the hopper area and pigeons were able to retrieve a mixture of grain and pellets. An infrared photobeam was projected across the opening of the hopper to detect head entry through disruption of the photobeam. All experimental events were controlled and recorded with a Pentium III-class computer (Dell, Austin, TX). A touchscreen and monitor located in the center of the front panel were covered with white cardboard.

Stimulus presentation and response detection were accomplished via an ARENA module constructed from acrylonitrile butadiene styrene (ABS) plastic. Each square plastic box was 12.9 cm wide and 3.5 cm high. In the center of the top plate of the module, a 3.0 cm diameter by 3.0 cm deep well made of polyvinyl chloride (PVC) plastic was inserted such that its opening was flush with the top plane of the module. An enclosed sensor circuit restricted the sensing field to the volume within the cup (described in more detail in Badelt and Blaisdell, 2008). The sensor was activated when a pigeon's beak (or any conductive mass) broke the top planar surface and entered the space within the well. The module also housed a radio-frequency transmitter/receiver by which it was networked to a PC computer in an adjacent room. The network allowed the module to receive command signals to change the status of the LEDs and to send detection events to the computer. Each module was equipped with six LEDs located in the bottom of the response area. Three of the diodes were positioned horizontally and the remaining diodes were positioned vertically facing up towards the opening of the well. The horizontal diodes were white LEDs and acted as the autoshaping CS or the instrumental discriminative stimulus in Experiment 1. The vertical diodes were multi-color LEDs that could produce a wide-range of colors by varying the intensity of their red, green and blue elements. These vertical diodes were used to produce the different colors that functioned as the instrumental discriminative stimuli in Experiments 2 and 3.

1.2. Procedure

1.2.1. Hopper training

On Day 1, prior to training, grain and pellets were placed on the floor of the chamber surrounding the opening to the hopper to facilitate approach and eating. Subjects received a 30-min session during which the houselight, hopperlight, and hopper were activated and remained active for the duration of the session. On Day 2, the houselight, hopperlight, and hopper were activated at the start of the session, but the hopper was lowered and the hopperlight extinguished after 5 cumulative seconds of head entry. Following a variable-time (VT) 15-s interval the hopper was again raised (hopperlight was activated whenever the hopper was in the raised position). Hopper presentations continued to occur in this manner throughout the session. Sessions ended after 50 hopper presentations or after 30 min had elapsed.

1.2.2. Autoshaping/instrumental shaping

Treatment consisted of six stages of seven sessions each, with a different manipulation occurring in each stage. In the first stage, the ARENA module was vertically positioned and located 11.51 cm from the hopper area (see Fig. 1). The well was lit with white light which served as the CS. During the intertrial interval (ITI) the well was not lit. On each trial the CS was presented for 10 s and terminated with 5-s access to reinforcement followed by a 90 ± 30 s variable-time (VT) ITI. Pecks into the well during a trial or ITI had no effect on the delivery of reinforcement but were recorded. Subjects received 35 trials per session. In the second stage, the module was placed in a horizontal position (see Fig. 1) and treatment was the same as in Stage 1. In Stage 3, trial duration was increased to 20 s and the ITI was increased proportionally, but only 18 trials were given in each session. In Stage 4, the intensity of the autoshaping cue was reduced by half. In Stage 5, an instrumental reinforcement contingency was in effect in addition to the Pavlovian contingency as described above. Each trial ended in reinforcement either after 20 s or when a peck was detected in the well. After Stage 5, the birds received a maximum of four sessions of instrumental shaping. Each shaping session was composed of three sections. At the onset of the session five trials identical to those in Stage 5 were given, after which shaping by successive approximations was implemented manually through the computer program that controlled trial events. During manual shaping the module remained lit for 15 min and reinforcement was delivered for 4-s following every observation of the required response. If the subject pecked the response area during shaping the module light extinguished and an ITI (VT-15 s) followed reinforcement. After the manual shaping section of a session, the birds completed the remaining trials as in Stage 5. Stage 6 began when subjects reliably pecked the lit module during manual shaping; all experimental parameters in Stage 6 were identical to Stage 5.

1.3. Results

Fig. 2 shows the mean peck rates during the CS and the ITI across all six stages of training as well as the proportion of trials with a response. The manipulations conducted during Stages 1–5 appear to have had no significant effect on either measure as indicated by the low proportion of trials with a peck and the lack of a difference in peck rates during the CS and the ITI. Only manual shaping of the instrumental response prior to Stage 6 had a dramatic effect on conditioned responding as indicated by the increase in the rate of responding during the CS and proportion of trials with a response.

An analysis of variance (ANOVA) with repeated measures conducted on mean proportion across Stages 1–6 yielded a main effect,
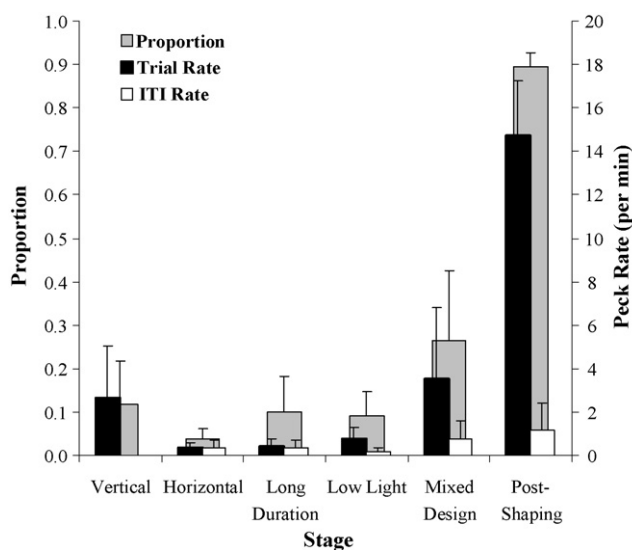
Fig. 2. The proportion of trials with a response (left axis) as a function of Stage, and mean response rate (right axis) during the ITI and trial in Experiment 1. Error bars represent standard error of the mean.
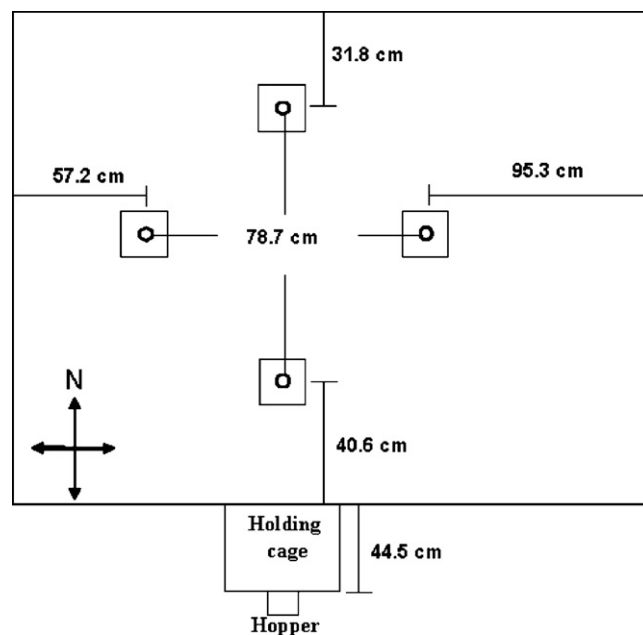

Fig. 3. A schematic representation of the test area and holding cage used in Experiments 2 and 3. The East–West orientation was used for Experiment 2 and the North–South orientation for Experiment 3. The grain and pellet dispenser is labeled 'hopper', and is located at the South end of the holding cage. Figure not drawn to scale.

$F_{(5, 20)} = 32.47$, $p < .01$. A post hoc analysis using Tukey's honestly significant difference (HSD) statistical test found mean proportion in Stage 6 differed from the remaining Stages, $ps < .01$, but no other comparisons were significant. A similar repeated-measures ANOVA conducted on response rate with Response (ITI and Trial) and Stage (1–6) as repeated measures revealed a marginally significant main effect of Response, $F_{(1, 4)} = 7.52$, $p = .05$, a main effect of Stage, $F_{(5, 20)} = 12.07$, $p < .01$, and the interaction, $F_{(5, 20)} = 10.78$, $p < .01$. Tukey's HSD was used to isolate the source of the interaction and revealed that the Trial Rate from Stage 6 differed from the Trial Rate in each of the remaining Stages, $ps < .01$, and from the ITI Rate in each Stage, $ps < .01$, but no other differences were found. These analyses indicate that although the pigeons occasionally pecked the device during the CS and during the ITI in the first five stages of training, they failed to autoshape (i.e., no difference in the rate of pecks during the Trial and ITI). It was only after the introduction of a manual shaping procedure prior to Stage 6 that the pigeons came to reliably peck the device when lit.

Though we report a failure to observe autoshaping in the group-level data averaged across birds, two birds were observed to peck on at least 22% of trials within the first two autoshaping sessions. Furthermore, these pecks only occurred while the module was lit. Thus, two birds appeared to rapidly acquire an autoshaping response. By the end of the first stage of training, one additional bird pecked at the plastic surface surrounding the recessed well while the module was lit, and two other birds were orienting to the CS. Thus, four out five subjects showed some behavioral evidence of learning the CS–US relationship. One reason for the difficulty in establishing autoshaping with our procedure may reside with the nature of the response to the module. During shaping, we observed that pigeons made shallow pecks that did not make contact with the bottom of the well. The lack of a tactile component to the peck may have been unnatural for some subjects and thereby contribute to their reluctance towards vigorous responding. Future modifications of the ARENA modules may be necessary to increase its effectiveness. Nevertheless, all subjects were successfully instrumentally shaped to peck the module in a highly reliable manner.

2. Experiment 2

The results from Experiment 1 suggest that the ARENA module can reliably deliver a discriminable signal and is able to detect subject responses across a variety of procedural variations and across many days, thereby extending the initial observations in a single session by Badelt and Blaisdell (2008). We failed to find evidence of control by a Pavlovian autoshaping contingency within the limited set of parameters we used, though we did find responses to a module were supported by an instrumental contingency. Instrumental conditioning is an extremely useful procedure that serves as the base for a wide variety of experiments in animal learning and cognition. Thus, in Experiment 2, we introduced a second ARENA module to assess the ability of the ARENA system to coordinate a procedure involving two modules. In addition, we implemented a more complex procedure consisting of a simultaneous visual discrimination. Two modules were placed in the test area in an East–West orientation (see Fig. 3). During a trial, the modules displayed one of two colors, with each module displaying a different color. For example, the East module might display red while the West module displayed green. Each color appeared equally often at each location within each session. One of the colors was designated as an S+ and pecks to that module were reinforced, while pecks to the module display the other color (S−) were not reinforced.

2.1. Method

2.1.1. Subjects

Three white Carneaux pigeons (C. livia; Double T Farm, IA) served as subjects. All subjects had previously been autoshaped in a touch-screen apparatus. Subjects had previously received manual shaping in ARENA and subsequently learned to respond for reward in the presence of a lit module. The remaining details are as in Experiment 1.

2.1.2. Apparatus

Experimental manipulations took place in an anechoic room with a test area of 2.3 m × 1.5 m. The walls of the room were covered in white sound insulating tiles. The East wall of the room had a full-sized door. On the South wall, there was an automated door and an access hole for wires connecting to a PC in an adjacent room. The

automated door which separated the test area from the holding cage was 31.8 cm high and 16.7 cm wide. The bottom of the automated door was 3.8 cm above the floor, and located 78.0 cm from the West wall and 112.0 cm from the East wall. Illumination was provided by four 25-W incandescent lights mounted on shower rods positioned 10.2 cm below the ceiling. The lights faced upward to provide indirect lighting. A white noise generator elevated 109.2 cm from the floor and centrally located above the automated door delivered an oscillating 56–63 db (A) background noise. Two Harman-Kardon multimedia speakers were mounted at the same height. A Sony SSC-DC374 color video camera with wide-angle lens was mounted on the ceiling 2.4 m above the floor of the test area and allowed for viewing and recording experimental procedures in the adjacent room.

This experiment used two ARENA modules of the type described in Experiment 1. The modules were arranged in an East to West orientation (see Fig. 3). The modules displayed one of four colors: red, blue, green, and yellow (see Section 2.2 for details). A hopper like that described in Experiment 1 was mounted on the rear wall of a holding cage with the hopper opening 10 cm above the floor of the cage. The holding cage was located on the other side of the automatic door from the test area. It measured 35.6 × 36.8 × 30.5 cm, was made of white acrylic, and was illuminated by a 28-V Med Associates houselight mounted to the ceiling.

2.2. Procedure

2.2.1. First discrimination

Pigeons received discrimination training with the red, blue, and green lights (each pigeon only received two of the three colors). The color assignments were counterbalanced across subjects so that each color equally often served as an S+ and an S−. On each trial, one module displayed the S+ color while the other module displayed the S− color. Within each session, each module displayed the S+ on half of the trials and the S− on the remaining trials in random order (i.e., the S+ occurred equally often within a session in the West and East modules). Responses at the module that displayed the S+ were reinforced according to a gradually increasing reinforcement schedule, while responses at the module that displayed the S− were neither reinforced nor punished. On reinforced trials, the hopper, houselight, and hopperlight were activated. In addition, a 0.5-s digital noise (windows xp xclamation.wav, 69 db [A-scale]) was played concurrently with the raising of the hopper. The hopper remained in the raised position until the infrared beam projected across the hopper opening was broken by the bird's head after which the hopper remained up for 3 s after which the ITI started. If no head entry was detected after 120 s following hopper onset, the hopper became inactive, the houselight and hopperlight extinguished, and the ITI started. Initially reinforcement was delivered on a fixed-ratio 1 schedule of reinforcement, followed by a shift to a variable-ratio 3 (VR-3), and then a VR-5 schedule. Each increase occurred after the subject performed with an accuracy score above 80%. Accuracy was calculated by dividing the number of trials that ended with a correct response by the total number of trials in the session. Trials ended with reinforcement or after 60 s from the onset of the stimulus, whichever came first. Trials that ended with a correct response were followed by a 10-s ITI, while trials that timed out led to a 70-s ITI. During the ITI the module lights were extinguished. Pecks emitted during an ITI were recorded but had no consequences. We calculated accuracy, discrimination ratios (S+ and S−), and response rates (ITI and Trial) for each session. The discrimination ratios were calculated as responses to either the S+ or S− divided by responses to both the S+ and S−. Response rates were calculated by dividing summed responses during either the ITI or Trial by the total duration of ITIs or Trials, respectively. Response rates per minute were then calculated. Whereas accuracy indicates the bird's general per-

formance during a session, the discrimination ratios and response rates give more information about performance during a trial. A session ended either after the subject completed 100 trials or after 60 min, whichever came first.

2.2.2. Second discrimination

After completing the first discrimination by achieving accuracy above 80% on two consecutive sessions with a VR-5 schedule of reinforcement, subjects received a second discrimination involving new colors: the unused color from the first discrimination and yellow. The assignment of color to S+ and S− was counterbalanced across birds. All procedural details were as described for the first discrimination.

2.3. Results

2.3.1. First discrimination

The top panel of Fig. 4 shows that the mean discrimination ratio increased across reinforcement schedules from the first block to the last. To test whether performance reflected a significant preference for the S+, we conducted *t*-tests for single means to assess discrimination ratios against a value of 0.5 which reflects no difference in peck rates to the S+ and S−. This test revealed no difference in Block 1, $t(3) = 2.19$, $p > .05$, but a discrimination ratio reliably greater than 0.5 for Block 2, $t(3) = 39.85$, $p < .001$. Tests were not possible for Block
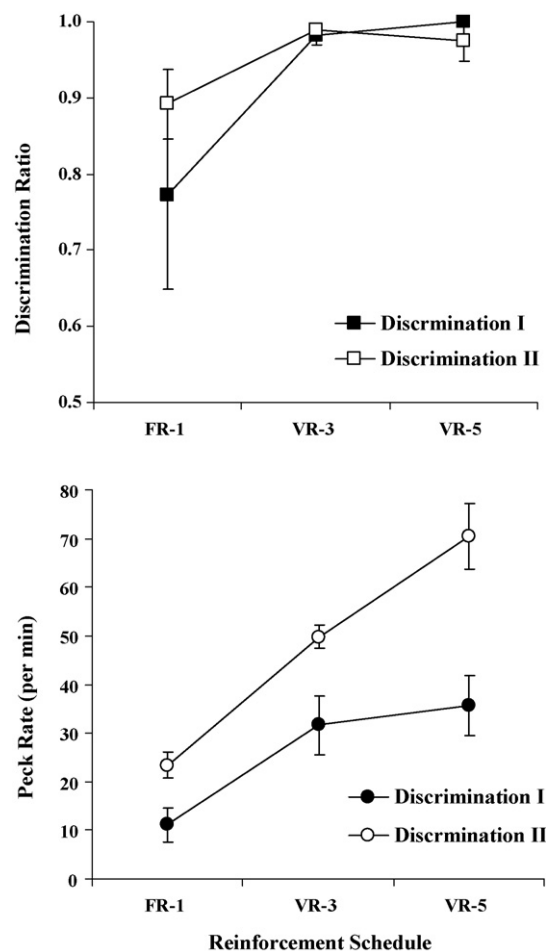

Fig. 4. Top panel: Mean discrimination ratios from the simultaneous discrimination of Experiment 2 as a function of reinforcement schedule. Bottom panel: Mean response rate at the S+ as a function of reinforcement schedule. The filled symbols indicate data from the initial discrimination, while the empty symbols are from the second discrimination. Error bars represent standard error of the mean.

3 as perfect performance by each subject produced a discrimination ratio of 1 with no variance; however, no statistics are required to confirm self-evident perfect performance. We conducted a similar *t*-test to assess biases to respond based on module location (East or West) and found no bias in any of the blocks, $ts(3) < 1.55$, $ps > .05$.

Mean response rate at the S+ increased across blocks (Fig. 4, bottom panel), while mean response rate at the S− decreased from the first block ($M = 2.15$, SD = 1.65) to the last ($M = 0.09$, SD = 0.16). An ANOVA with repeated measures conducted on mean response rate with Response (S+ and S−) and Block (1–3) as repeated measures confirmed a main effect of Response, $F(1, 2) = 30.77$, $p < .05$ and Block, $F(2, 4) = 16.42$, $p < .05$, and revealed a Response × Block interaction, $F(2, 4) = 13.91$, $p < .05$. Tukey's HSD revealed that the S+ response rate in Block 1 differed from Blocks 2 and 3, $ps < .05$. Additionally, response rates at the S+ differed in Blocks 2 and 3 from response rates at the S− in each Block (1–3), $ps < .01$.

2.3.2. Second discrimination

Subjects acquired the second discrimination in fewer sessions ($M = 6$, SD = 0) than they did the first ($M = 8$, SD = 1.73), though this difference was not significant, $t(2) = 2.0$, $p > .05$. Fig. 4 also shows mean discrimination ratios (top panel) and response rates to the discriminative stimuli (bottom panel) as a function of reinforcement schedule for the second discrimination. *t*-Tests for single means revealed that the discrimination ratio in each block was significantly above 0.5, $ts(3) > 8.56$, $ps < .05$. A similar *t*-test on responses to the position (East–West) of the module found no difference from 0.5, $ts(3) < 2.78$, $ps > .05$, indicating no spatial biases among the birds. As with the first discrimination, the response rate at the S+ increased dramatically across block (Fig. 4, bottom panel) while mean response rate at the S− showed only a marginal increase from the first ($M = .90$, SD = 1.57) to last block ($M = 2.02$, SD = 2.50). An ANOVA with repeated measures conducted on mean response rates with Response (S+ and S−) and Block (1–3) as the repeated measures revealed a main effect of Response, $F(1, 2) = 82.72$, $p < .05$, Block, $F(2, 4) = 44.44$, $p < .01$ and the interaction, $F(2, 4) = 44.68$, $p < .01$. Tukey's HSD revealed response rates at the S+ differed across each Block, $ps < .01$, and in each Block response rates at the S+ differed from the S−, $ps < .05$, no other differences were found.

In Fig. 4, response rates during the S+ showed a steeper function across blocks in the second than first discrimination. An ANOVA with repeated measures conducted on response rate during the S+ with Discrimination (1–2) and Block (1–3) as the repeated measures revealed a main effect of Block $F(2, 4) = 34.32$, $p < .01$, and the interaction, $F(1, 2) = 9.08$, $p < .05$. Tukey's HSD revealed that the source of the interaction is the difference in response rates at the S+ between Discrimination 1 and 2 during Block 3, $p < .01$.

The results from this experiment indicate that subjects were able to use color of the module light as a discriminative signal for appropriate responding. Subjects initially responded to both devices but over time learned to discriminate the S+ from the S− colors. Subjects did not show a bias for module positions. These results extend the reliability of ARENA to a situation involving two modules.

3. Experiment 3

In a simultaneous discrimination task, the S+ and S− are both present simultaneously on each trial. Like successive discrimination tasks (e.g., the go/no-go procedure), successful performance requires learning which stimuli signal a response-reward contingency and which signal a response-no-reward contingency. A conditional discrimination is a more complicated procedure because successful performance requires learning what type of response is appropriate for each stimulus. For example, both ARENA

modules may be lit with the same color on each trial but different colors across trials (e.g., both red on trial 1, both blue on trial 2, etc.). Furthermore, pecking one of the two modules (e.g., the North module if they are arranged in a North–South orientation, see Fig. 3) may be rewarded in the presence of some colors, but pecking the South module may be rewarded in the presence of other colors. This conditional discrimination requires attending to both what color is present on a trial and what response (North or South) is appropriate in the presence of that specific color. This experiment evaluated the ability of pigeons to solve this type of conditional discrimination in ARENA. On any given trial, one of six colors was illuminated in both devices concurrently, and signaled that a response at one of the spatial positions would be reinforced (S+) while a response at the other position would not (S−). Subjects needed to learn the conditional relation between the color and the position associated with reinforcement. After learning the conditional relationship, the color-location assignments were reversed to assess the flexibility of the subjects to re-adapt.

3.1. Method

3.1.1. Subjects

Four white Carneaux pigeons (*C. livia*; Double T Farm, IA) served as subjects. All subjects had previously been autoshaped in a touchscreen apparatus and served in a spatial search task on the touchscreen. Subjects had previously received manual shaping in ARENA and subsequently learned to respond for reward in the presence of a lit module. The remaining details are as in Experiment 1.

3.1.2. Apparatus

Same as in Experiment 2, except that the colors displayed in the response area included light green, light blue, yellow, orange, pink, and maroon, and the modules were placed in a North–South orientation (Fig. 3). Two ARENA modules of the type described in Experiment 1 were used in this experiment. Each module was placed 96.6 cm from the East wall of the test area and 134.7 cm from the West wall of the test area. The North module was 119.3 cm and the South module was 40.6 cm from the wall that contained the holding cage and hopper. This meant that the North module was 163.8 cm and the South module was 85.1 cm from the food hopper. The modules were separated by 78.7 cm from center to center.

3.2. Procedure

3.2.1. Conditional discrimination

On each trial, one of six colors was displayed in both modules concurrently. The color signaled that responses at one (S+) of the two locations were reinforced while responses at the other location (S−) were never reinforced. Out of the eight possible colors, six colors were selected and divided into two sets of three colors: Sets A and B. Stimuli in Set A signaled reinforcement at one location (e.g., North) while Stimuli in Set B signaled reinforcement at the other location (e.g., South). The assignment of color to Sets A and B were counterbalanced across subject. Initially trial outcomes were delivered on an FR-1 schedule. The reinforcement schedule increased from an FR-1 to a VR-3 and finally to a VR-4 each time after one session with an accuracy above 80%. Module pecks were counted cumulatively so that whichever module (S+ or S−) accumulated the criterion number of pecks first determined trial outcome. Trial order was randomly determined with the constraint that there was an equal number of each trial type in each session. Each trial was separated by a VT 15-s ITI. The modules were unlit during the ITI. Pecks emitted during an ITI were recorded but had no consequence. We calculated accuracy, discrimination ratio for the S+ and

S−, and response rates (ITI and Trial) for each session. A session ended either after the subject completed 100 trials or after 60 min, whichever came first.

3.2.2. Discrimination reversal

After completing two consecutive sessions on a VR-4 schedule of reinforcement above 80% accuracy, the subject progressed to the reversal phase during which the assignments of stimuli to reinforced location were reversed. For example, if the stimuli in Set A had been assigned the North module as S+, they were now assigned the South module as S+, and likewise for the stimuli in Set B. One bird initially never responded correctly in the presence of one of the colors and so this subject received two sessions of 20 trials with just this color to correct the problem. The subject then resumed normal training.

3.3. Results

Subjects took a Mean of 14.25 (SD = 0.96) sessions on the initial discrimination to achieve above 80% accuracy on a VR-4 schedule. Fig. 5 (top panel) shows that mean discrimination ratios were greater than 0.5 across each reinforcement schedule. These observations were confirmed by a *t*-test for single means which assessed group performance against a discrimination ratio of 0.5. This test revealed differences in each Block of reinforcement schedule $ts(4) > 8.91$, $ps < .01$. A similar *t*-test was

Fig. 5. Top panel: Mean discrimination ratios from the conditional discrimination of Experiment 3 as a function of reinforcement schedule. Bottom panel: Mean response rate at the S+ as a function of reinforcement schedule. The filled symbols indicate data from the initial conditional discrimination and the empty symbols are from the reversal. Error bars represent standard error of the mean.

conducted with the discrimination ratio for module preference and showed that none of the Blocks approached significance, $ts(4) < 3.07$, $ps > .05$.

The bottom panel of Fig. 5 shows that response rate at the S+ increased across schedules. Response rate at the S−, however, showed little change from the first block ($M = 3.20$, SD = 1.84) to last block ($M = 4.00$, SD = 2.15). These observations were confirmed by an ANOVA with repeated measures calculated on mean response rates with Response (S+ and S−) and Block (1–3) as the repeated measures. This analysis revealed main effects of Response, $F(1, 3) = 98.54$, $p < .01$, Block, $F(2, 6) = 27.42$, $p < 0.01$, and the interaction, $F(2, 6) = 48.31$, $p < 0.01$. Tukey's HSD revealed that the response rate at the S+ during Block 1 was different than Blocks 2 and 3, $ps < 0.01$, while Blocks 2 and 3 did not differ from one another. Response rates at the S− did not differ from one another across Block, but did differ from response rates at the S+ during Blocks 2 and 3, $ps < 0.01$.

Subjects took a Mean of 17.50 (SD = 0.58) sessions to reach above 80% accuracy during the reversal, which is greater than the mean number of sessions to learn the conditional discrimination (Mean = 14.25, SD = 0.96). A *t*-test for dependent samples conducted on the number of sessions to reach criterion revealed a difference between the conditional discrimination and reversal, $t(4) = 4.33$, $p < 0.05$. As in the original discrimination, mean discrimination ratios were above 0.5 in each block of reinforcement schedule indicating control by the reinforcement contingency, $ts(4) > 4.86$, $ps < 0.05$. Discrimination ratios for module preferences revealed no differences in any of the Blocks, $ts(4) < 1.63$, $ps > 0.05$.

The bottom panel of Fig. 5 also shows that during reversal training the response rate at the S+ increased across Block. The reverse pattern was observed in the response rate at the S− from the first ($M = 6.57$, SD = 3.67) to last block ($M = 4.43$, SD = 2.67). These observations were confirmed by an ANOVA with repeated measures calculated on mean response rates with Response (S+ and S−) and Block (1–3) as the repeated measures which revealed a main effect of Response, $F(1, 3) = 275.42$, $p < .01$, Block, $F(2, 6) = 37.66$, $p < .01$, and the interaction, $F(2, 6) = 38.26$, $p < .01$. Tukey's HSD found the response rate at the S+ during Block 1 to be different than Blocks 2 and 3, $ps < 0.01$, but that Blocks 2 and 3 did not differ from one another. Response rates at the S− did not differ from one another across Block, but did differ from response rates at the S+ during Blocks 2 and 3, $ps < 0.01$. These findings replicate the response rate changes in the conditional discrimination and reveal the source of the interaction was due to an increase in response rate at the S+ while the response rate at the S− showed little variation.

The conditional discrimination of Experiment 3 was more complicated than the simple discrimination of Experiment 2. In the simple discrimination, each color signaled that a response would be rewarded or not rewarded. In the conditional discrimination, each color signaled which of two spatial positions would be reward for a response. These results show the utility of using ARENA for rule-based (i.e., conditional) learning tasks.

4. General discussion

The experiments reported here demonstrate the utility of ARENA as an alternative to conventional open-field procedures. Although birds failed to acquire a pure autoshaping response in ARENA, they readily acquired a simple instrumental response (Experiment 1), a simple discrimination (Experiment 2), and a more complex conditional discrimination (Experiment 3). The patterns of behavior shown in ARENA were comparable to those found in similar experiments using conventional open-field procedures and automated systems like operant chambers equipped with either keylights or a touchscreen (e.g., Brodigan and Peterson, 1976; Cole and Honig, 1994; Emmerton, 2001; Kendall, 1983; Nevin, 1967; Perkins et al., 1976; Reynolds and Limpo, 1969).
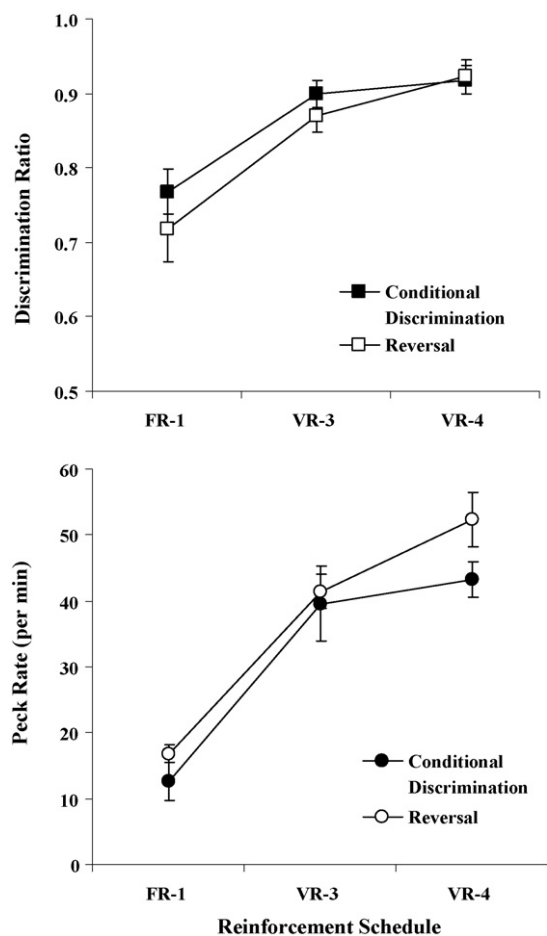
Although behaviors in the operant box have been likened to foraging behavior, the structural properties of the operant box require rather unnatural behavior for pigeons. For example, the subjects' movements within the operant chamber are extremely limited, and responses directed at a vertical surface do not approximate natural pecking behavior during foraging despite demonstrated control over the form of the CR by the nature of the reinforcer (e.g., Brown and Jenkins, 1968). The automated open-field environment provided by ARENA, however, allows the subject to emit normal movements engaged during foraging behavior, such as approaching a goal from multiple directions. Moreover, the ARENA modules can be placed horizontally on the floor of the open field so that the subject can peck down into the well serving as the response location. Finally, modules may be placed at various locations to simulate changing food sources and availability without generating a preference for one module or location over another. While not a focus of the current studies, temporal (e.g., latency to respond) aspects of the response could also be recorded with this system, allowing the technology to be applied to a greater set of research questions (see Badelt and Blaisdell, 2008, for a discussion of ARENA's flexibility).

Although ARENA shares with conventional discrete-trial open-field procedures greater ecological validity, its operant nature obviates many of the drawbacks to such procedures. Whereas conventional open-field procedures and data collection tend to be laborious, error prone and subjective, and disruptive to the subject, ARENA suffers none of these shortcomings. Although ARENA awards the subject greater behavioral flexibility, the use of a proximity sensor for detection of a response provides an objective and reliable measure of behavior (Badelt and Blaisdell, 2008). Furthermore, the use of a single food hopper in a fixed location to dispense all reinforcement removes the concern over potential odor and visual cues unintentionally produced by having food located at the stimulus-response locations. In addition, the operant nature of ARENA allows for the collection of many more trials in a session as well as many more types of trials (e.g., counterbalancing, control manipulations, etc.). As a point of contrast, conventional discrete-trial open-field studies using rats and pigeons are typically limited to anywhere from 3 to 15 trials per session (e.g., Biegler and Morris, 1999; Blaisdell and Cook, 2005; Cheng, 1986, 1988; Spetch et al., 1997). In ARENA, we have collected data in an 8-h day from as many as six birds with 100-trial sessions each. This not only increases the speed at which subjects can progress through various stages of training and testing, it also allows for an increased number of control trials or alternate treatments that can be presented in a single session. As a result, more complex within-subject designs can be used (as in Experiments 2 and 3) which reduce the number of subjects required for a single study and increases its power.

Although ARENA shares with the Skinner box the benefits attributable to an operant procedure, as mentioned above it reduces the contrived nature of the Skinner box. Cook et al. (2004) directly compared the acquisition of a simultaneous discrimination in rats using either a conventional Skinner box with levers or a touchscreen-equipped operant chamber. They found that the group of rats trained to respond using a touchscreen learned the simultaneous discrimination faster than the group of rats responding using levers. They suggested that the difference in the rate of acquisition of the instrumental response was due to the touchscreen response being more similar to the rat's natural foraging behavior (e.g., scrabbling) than is a lever press response. Similar observations have been reported for pigeons when the response required better approximates natural foraging behavior, such as pecking down at a horizontal touchscreen (Wright et al., 1988) or into a gravel filled cup (Wright and Delius, 1994). Like the procedures used by Wright et al., ARENA requires a more natural downward oriented response rather than the forward response made at head height required by a conventional operant box. More importantly, the pigeon can move about the 3D environment of ARENA which is far more natural to a foraging pigeon than being constrained to the front of a 2D vertical surface as in conventional operant chambers with either key lights or a touchscreen. Thus, the ARENA setting more closely approximates the pigeon's natural foraging environment. Despite these advantages, the touchscreen operant box is still a useful technology for questions ARENA is not equipped to address, such as those involving complex visual displays (e.g., in scene and object perception) and moving stimuli and video content.

The wireless capabilities of the ARENA modules allow them to be arranged in almost any configuration or position, thereby adding a great deal of flexibility to their use. Results from Experiment 3, in particular, also confirm that a module placed nearer to the food hopper than another module does not negatively influence search behavior. Modules may be placed in various geometric configurations (e.g., circular, square, and linear), mounted on various surfaces (e.g., floor, wall, and platforms of various heights), and the spacing between modules can be manipulated. Likewise, the number of available options provided to the subject can be controlled by adding or removing individual modules. Modules may be placed in a conventional open field for use with human and non-human animals, and additionally, placed inside (e.g., classroom) or outside (e.g., courtyard) for use with humans. In addition, hopper sites at more locations (e.g., North, East, and West walls) throughout the test area (or at the modules themselves) may better approximate natural foraging conditions.

The development of a fully automated apparatus for the investigation of cognitive and behavioral processes in pigeons signals a significant advancement to open-field experiments. Pilot studies in our lab have also found the technology suitable for other subjects besides pigeons, such as rats and mice (Badelt and Blaisdell, 2008). As discussed by Badelt and Blaisdell (2008), Cook et al. (2004), and Walsh and Cummins (1976) technological advances in behavioral recording in terms of flexibility, reliability, and effectiveness apply not only to basic behavioral research, but may also advance research into the genetic and neural basis of behavior. Though both conventional preparations are important to verify the processes involved in spatial learning (the open field for its continuous search space and the operant box for its flexible display), ARENA provides an important new technology that combines the precision and efficiency of the operant box with the ecological validity of more traditional open-field preparations.

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