ODDITY LEARNING IN HONEYBEES (*APIS MELLIFERA*)

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In dedication to Shane Wilson-South.

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Introduction

"I know, indeed, that brutes do many things better than we do, but I am not surprised at it; for that, also, goes to prove that they act by force of nature and by springs, like a clock, which tells better what the hour is than our judgment can inform us" (Rene Descartes, 1646).

Historical Overview

Descartes attributed to humans both voluntary action and involuntary reflexes while to nonhuman animals only innate involuntary reflexes (1637). This idea persisted until Charles Darwin published his ideas on evolution in the two books, *On the Origin of Species* (1859) and *The Descent of Man* (1871), where he speculated about the mental continuity between humans and nonhuman animals. To provide evidence of mental continuity, George Romanes, Darwin's pupil and collaborator, published *Animal Intelligence* in 1892. Even though Romanes relied heavily on anecdotes and anthropomorphic interpretations, he nonetheless attempted to characterize the evolution of intelligent behavior. He provided observational accounts of both invertebrates (e.g., scorpions, bees, ants, mollusks, including oysters and octopuses) and vertebrates (e.g., fishes, reptiles, and mammals) with particular emphasis on cats, dogs, and elephants (Romanes, 1892).

Conwy Lloyd Morgan, Romanes' successor, was more critical than Romanes and rejected anecdotes and anthropomorphic interpretations in favor of empirical evidence. He conducted field experiments on reflexes with several species, including young chicks, ducklings, and dogs. Morgan is best known, however, for his cautionary statement, "In no case may we interpret an action as the outcome of the exercise of a higher psychical faculty if it can be interpreted as the outcome of the exercise of one which stands lower in

the psychological scale" (Morgan, 1894). This statement became known as Morgan's Canon, a restatement of Occam's Razor in psychological terms.

Also, in response to Romanes' anthropomorphism and lack of experimentation, Edward Thorndike brought the study of animal intelligence into the laboratory. Thorndike viewed the anecdotal observations of animal intelligence as "eulogies," that is, an emphasis on the unusual and the marvelous animal behavior rather than on the normal and the unremarkable (Thorndike, 1898). To remedy this situation he built "puzzle boxes" from which experimental subjects, such as cats, dogs, and young chicks, had to escape. He then measured escape latencies in a series of trials with each species of subject, all of which exhibited the same results. The escape latencies gradually decreased over trials. The subjects never exhibited spontaneous or insightful learning as would be indicated by an immediate and sharp decrease in escape time. From these experiments, Thorndike established the Laws of Effect and Exercise, two cornerstones of associative learning (1911). The rigorous experimental methods Thorndike used to study animal learning became the foundation for the study of instrumental conditioning, later called operant conditioning by Skinner (1938), a procedure where responses are followed by reward or punishment.

At the same time Thorndike was conducting his experiments, Ivan Pavlov was conducting experiments on the digestive reflexes of dogs (Pavlov, 1927). Accidentally, he discovered the conditioned reflex. When he noticed his dogs salivated before any food had been placed in their mouths, he concluded that a novel stimulus paired with an unconditioned reflex elicited a conditioned reflex. In his subsequent work, he attempted to characterize the parameters necessary to produce conditioned reflexes. Pavlov's work later became the foundation for classical or Pavlovian conditioning. Both Pavlovian and instrumental conditioning procedures provide powerful tools for the laboratory study of learning.

Invertebrate Learning

Initially, research was conducted on a wide variety of species to determine the range of species that could learn. However, once it was established that all species likely could learn, subsequent research on learning focused primarily on the rat and pigeon (Beach, 1950; Bitterman, 2006). Both the rat and the pigeon were considered ideal candidates for research subjects in studies of learning because they are abundant, cheap, hardy, and easily reared for the laboratory (Beach, 1950; Bitterman, 2006). The work with these species served to facilitate the development of general learning principles, assumed to apply to all species. Later, technological advances in cellular neuroscience facilitated a renewed interest in the learning of other species, including invertebrates. Invertebrates such as gastropods (e.g., aplysia) and crustaceans (e.g., crayfish) proved ideal for studies of the biological basis for learning because they have relatively simple nervous systems with very large and fast neurons (Corning, Dyal, & Willows, 1973).

Interestingly, while gastropods and crustaceans make ideal subjects for electrophysiological experiments, they are not ideal subjects for learning experiments because of their limited sensory and motor repertoires. For this reason, invertebrate learning studies have focused primarily on cephalopods (e.g., octopuses) and insects (e.g., honeybees). These species are not without limitations for learning work, although, arguably, the octopus presents more methodological problems than the honeybee (see: Corning, Dyal, & Willows, 1975; Bitterman, 1975; Boal, 1996). Octopuses are not hardy in the lab and require special housing and proper handling. Conversely, honeybees are easier to maintain for laboratory study. Furthermore, honeybees have a range of sensory, motor, and motivational capabilities that make them ideal subjects for learning research.

Karl von Frisch (1950) conducted experiments to determine the sensory capacities of the honeybee. He found that honeybees have color vision similar to that of humans, although they do not detect longer wavelengths (e.g., red) and do detect shorter wavelengths (e.g., ultraviolet). Honeybees have chemoreceptors on their antennae and tarsi (von Frisch, 1950; Scheiner, Page, & Erber, 2001) which are important for kin

discrimination, recognition of floral odors, and perception of pheromones (Robertson & Wanner, 2006). Honeybees also detect magnetic fields (Gould, Kirschvink, & Deffeyes, 1978; Walker & Bitterman, 1985) as well as vibration and touch (Kirchner, 1993; Tautz & Rohrseitz, 1998; Rohrseitz & Tautz, 1999; Nieh & Tautz, 2000). The motor capabilities of honeybees include flying, walking, oscillating antennae, as well as stinger and proboscis extension, all of which provide a number of response systems for learning experiments. In addition, motivated for nectar (sugar), foraging honeybees will consistently return to a high concentration of sugar solution and will do so without satiating since they are able to regurgitate their food at the hive in order to make room for more.

In early experiments on learning, researchers were mainly interested in exploring basic learning phenomena in the honeybee using analogs of vertebrate learning experiments. Two procedures were developed for studying learning in honeybees, the proboscis extension and the free-flying techniques. The proboscis extension technique uses foraging honeybees that are caught and then restrained, allowing only minimal head movement. When a bee's antennae are touched with sucrose, the proboscis extends, a response referred to as an unconditioned reflex. This unconditioned reflex can be paired with a novel stimulus such as odor to elicit a conditioned reflex of proboscis extension. Traditionally, proboscis extension has been used to explore basic Pavlovian conditioning and has provided much evidence that the conditioning of honeybees is similar to vertebrates (Bitterman, Menzel, Fietz, & Schäfer, 1983; Batson, Hoban, & Bitterman, 1992; Couvillon, Hsiung, Cooke, & Bitterman, 2005). The proboscis extension technique has been useful for the study of Pavlovian conditioning, but it does have limitations. The experimental bees must be restrained which permits satiation, and only odors or touch applied to the antennae can serve as the conditioned stimuli. To date, visual stimuli are not effective as conditioned stimuli for a conditioned response of proboscis extension. Due to these limitations, the free-flying technique is generally regarded as more powerful for conducting learning experiments with honeybees.

The free-flying technique uses an individual foraging honeybee trained to visit a laboratory window or table to feed on artificial flowers. The bee must learn about the flower's color or scent in order to receive a high concentration of sucrose reward. After it feeds, the bee is free to return to the hive and then again to the laboratory. This method has been used to study acquisition, extinction, parameters of reward, and choice discrimination in honeybees (Couvillon & Bitterman, 1985; Couvillon & Bitterman, 1988; Couvillon, Lee, & Bitterman, 1991; Bitterman, 1996), and, as with the proboscis extension technique, the results are similar to those of vertebrates.

Such similarities in general learning phenomena are remarkable given that vertebrates and invertebrates are believed to have shared a common ancestor some half a billion years ago. Furthermore, the brain structures of honeybees are very different than those of vertebrates. The similarities might make sense, however, if the biological mechanisms of learning occur at the cellular level (Kandel & Hawkins, 1992). Albeit, convergent evolution is perhaps more likely, that is, "different phenomena may be produced by the same processes, and what appear to be identical phenomena may be produced by different processes" (Bitterman, 1975).

Associative Versus Cognitive Explanations

In the spirit of Thorndike and Pavlov, the work with honeybees has concentrated on associative learning principles in order to facilitate comparison with vertebrates. Thorndike and Pavlov's associative approach, in fact, characterized the development of the field of animal learning with the exception of some early work with primates on insight, problem-solving, and language. However, there was work with other vertebrate animals that hinted at cognitive capacities beyond associative learning.

Although later discredited, Wolfgang Kohler (1918) conducted experiments on chickens and chimpanzees and proposed a cognitive rather than an associative explanation of discrimination learning. In his experiments, subjects were trained with reward to choose a light gray stimulus (S+) instead of a darker gray stimulus (S-) drawn

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from stimuli on a black to white continuum. After training, a transposition test was then presented to the subjects in which they had to choose between the original gray stimulus (S+) and a new, even lighter, stimulus along the black to white continuum. He observed that the subjects chose the lighter stimulus twice as often, leading him to conclude that the chickens were capable of learning "togetherness" or "relations" (Kohler, 1918).

Later, in direct opposition to Kohler's (1918) relational account of discrimination learning, Kenneth Spence (1936) proposed the first formal theory of discrimination learning. He claimed that Kohler's experimental subjects would consistently choose the lighter stimulus, not based on the ability to learn about relations, but on the absolute properties of the stimuli. Spence believed animals acquired an excitatory (E) tendency to choose a rewarded stimulus and an inhibitory (I) tendency to avoid a nonrewarded stimulus; both tendencies generalize to other stimuli on the same stimulus continuum and performance is determined by E - I. Spence (1936) concluded that, at least in theory, relational learning was not necessary to account for Kohler's results.

Spence's theory of discrimination learning provided further support for associative explanations of animal learning. Not until Edward Tolman, a major figure in the "cognitive revolution," published a paper in 1948 theorizing about the spatial mapping abilities of rats was there a renewed interest in cognitive explanations of learning. In his seminal paper, he conducted a series of experiments on maze learning in rats and concluded that they had learned to correctly navigate, not on the basis of associative learning responses, but rather on the basis of "cognitive maps." Tolman's idea of "cognitive maps" was later shown to be an inadequate explanation of the rats' maze learning abilities. Instead of relying on "cognitive maps" the rats were relying on extra-maze cues such as visible landmarks that could be seen from inside the maze and could serve to guide navigation. Regardless, his ideas signaled a change in perspective from an associative explanation to a cognitive explanation of learning.

Tolman's work on spatial learning in rats inspired a lot of research on memory using mazes, particularly the radial arm maze (Olton & Samuelson, 1976). Traditionally developed for the use of rats, a typical radial arm maze has 8 protruding arms from a

center circle. In studies of spatial learning, all the arms are baited in a trial, and, in order to receive food reward, the rat must visit all of the arms. An error in memory is a revisit to an empty arm. Rats were able to successfully navigate the maze with limited revisits, suggesting they have spatial short-term memory.

Just as Tolman's ideas inspired work using the radial arm maze to explore rats' spatial memory, his ideas also inspired work using the matching-to-sample (MTS) procedure to explore pigeons' short-term memory (Blough, 1959). In a typical MTS procedure, a sample stimulus is presented for a predetermined time, then there is a simultaneous presentation (or sometimes a successive presentation) of two choice stimuli, one different from and one the same as the sample. In order for a subject to receive a food reward, a correct choice must be made to the choice-stimulus that is the same as the sample stimulus. This procedure and its variation, nonmatching-to-sample (NMTS), have also proven useful for studying concept learning. (Note that the term concept is used throughout this thesis to indicate learning that may not be explained with basic principles of association.)

Along with the MTS and NMTS procedures, concept learning is explored using a same/different (SD) procedure (Wright & Katz, 2006). Typically in a SD procedure, pairs of stimuli are judged as same or different by a subject's choice of corresponding "same" or "different" response areas. In SD, MTS, and NMTS procedures, concept learning is assessed by transfer tests to novel stimuli, with good 1st trial performance indicating positive transfer (Thomas & Noble 1988). Recently, using SD, MTS, and NMTS procedures, evidence of concept learning has emerged in baboons, rhesus and capuchin monkeys, parrots, pigeons, and possibly even in honeybees (Wright & Katz, 2006; Chittka & Jenson, 2011). Concept learning seems to be a general vertebrate phenomenon, warranting an exploration of whether concept learning is unique to vertebrates, or whether invertebrates, like the honeybee, are capable of learning concepts as well.

Honeybee Concept Learning

To date, there have only been four published studies exploring whether honeybees are capable of learning concepts. Honeybees have been trained in discrimination problems that include symmetry/asymmetry (Giurfa, Eichmann, & Menzel, 1996), same/different (Giurfa, Zhang, Jenett, Menzel, & Srinivasan, 2001), above/ below (Avarguès-Weber, Dyer, & Giurfa, 2011), and a combination of above/below, left/right and difference (Avarguès-Weber, Dyer, Combe, & Giurfa, 2012). Each yielded significant results using methodological paradigms similar to those used with vertebrates.

Giurfa et al. (1996) conducted an experiment to determine whether honeybees could discriminate symmetry. Honeybees were presented with a succession of eight triads; each contained three different stimuli presented simultaneously. Depending on an individual bee's assignment, the correct choice was the symmetrical stimulus rather than the different asymmetrical stimuli (A+ B- C-), or the asymmetrical stimulus rather than the different symmetrical stimuli. Unrewarded transfer trials were interspersed among training trials. The bees' performance on the transfer trials was similar to that on the training trials, leading the authors to conclude that the bees had learned to abstract the cue "symmetry." Whether the honeybees actually learned "symmetry," however, is debatable given that transfer of learning is only indicative of concept learning if the 1st trial performance is above chance (Thomas & Noble, 1988) and first trial data were not provided.

To validate and further explore concept learning in honeybees, Avarguès-Weber et al. (2011) conducted an experiment using a MTS procedure to assess whether honeybees could master a conceptual spatial relation such as above and below. After training trials, the bees were presented with an unrewarded transfer test to determine whether concept learning had taken place. The results showed positive transfer, however, it could be argued that the bees did not learn above/below concepts, but merely solved a simple spatial discrimination problem.

Giurfa et al. (2001) performed another experiment using MTS and NMTS procedures to assess "same" and "different" concepts. Results of the honeybees' performance in both training and transfer tests were significantly better than chance, leading the authors to suggest the bees were learning concepts. Although, given the relatively large number of training trials (60 trials) that each bee received, it is possible the honeybees had just memorized all possible responses (only four responses), and thus learned a purely associative stimulus-response strategy. In addition, 1st trial transfer performance was not measured, leaving it unclear whether the honeybees had learned concepts or if generalization to new stimuli took place during the multiple transfer trials.

Additional experiments of simultaneous above-below, left/right, and difference concept learning in honeybees by Avarguès-Weber et al. (2012) yielded results similar to the studies mentioned above. Thus, Avarguès-Weber et al (2012) suggest, based on the honeybees' significantly better than chance performance and successful transfer, that honeybees are able to abstract concepts. This conclusion came only after the authors claimed to have excluded alternative explanations like template matching, perceptual generalizations, difference in centers of gravity, broad orientation cues, and pixel-by-pixel comparison. Upon closer inspection, however, the transfer tests did not meet the criterion to demonstrate transfer of learning as established by Thomas and Noble (1988).

The results of all four studies, taken together, "hint" at the possibility that honeybees may learn "concepts." These studies, although flawed, also suggest that concept learning is not unique to vertebrates. Of particular interest here is oddity learning, generally regarded as one of the ways to study concept learning in nonhuman animals (Robinson, 1933; Roitberg & Franz, 2004; Hille, Dehnhardt, & Mauck, 2006).

Oddity Learning

Robinson (1933) is one of the first researchers to develop and present an animal subject (e.g., macaque monkey) with an oddity problem. In order to test whether the animal was capable of responding based on the relationship between stimuli, he designed an oddity experiment in which one odd and two identical nonodd stimuli were simultaneously presented to a subject. To ensure that the successful solution of the problem was not due to cues such as position, color, or form, Robinson (1933) presented two kinds of trials: with one stimulus odd (A+ B- B-) on half the trials, the other stimulus odd (B+ A- A-) on the next. If the two kinds of trials are intermixed irregularly, successful solution might suggest a subject had learned to abstract the "odd" concept. Results of this early experiment using a single monkey indicated that an animal might be able to learn things other than associative responses. Although, since there were only six possible arrangements of stimuli (e.g., ABB, BAA, ABA, BAB, AAB, BBA), it is possible the animal had just memorized all possible combinations.

Over the next decades many different animals' oddity learning abilities were sampled using procedures similar to those used by Robinson (1933) to determine if oddity was a general learning phenomenon. Subsequent oddity problems were given to chimpanzees (Nissen & McCulloch, 1937), monkeys (Strong & Hedges, 1966), rats (Wodinsky & Bitterman, 1953), canaries (Pastore, 1954), and cats (Boyd & Warren, 1957; Warren, 1960) all of which were able to correctly solve an oddity learning problem. Although, cats were able to solve the oddity problem in some studies (Boyd & Warren, 1957; Warren, 1960), there was also evidence for cats' inability to learn the oddity problem (Strong & Hedges, 1966).

The success of nonhuman animals in oddity problems spurred interest in whether young children, ages four to seven years of age, also could solve an oddity problem. Children have been presented with the oddity problem with verbal instructions (Lipsitt & Serunian, 1963; Gollin & Shirk, 1966) as well as with nonverbal instructions (Overman, Bachevalier, Miller, & Moore, 1996) in order to simulate the oddity experiments using

nonhuman animal subjects. Experiments with child subjects provided evidence that a child's proficiency on an oddity problem is correlated with age as well as type of instruction given. Interestingly, children presented with an oddity problem and given only nonverbal instructions were unable to solve the problem with adult level proficiency (zero incorrect responses) until the age of seven (Overman et al, 1996).

At the same time that researchers were exploring oddity learning abilities in different species, they were also exploring the effects of manipulating the number of incorrect alternatives and placement of reward. The rate of learning in an oddity task has been shown to be facilitated by an increase in the number of incorrect alternatives in chimpanzees (Nissen & McCulloch, 1937), canaries (Pastore, 1954), and pigeons (Zentall, Hogan, Edwards, & Hearst, 1980). Other studies have manipulated placement of reward to determine whether there is an effect on learning if reward is presented across trials on the odd stimulus in all three positions, left, middle, and right (e.g., ABB, ABA, AAB, BAA, BAB, BBA), or if reward is presented across trials on only the left or the right stimulus (ABB, BAA, BBA, AAB). The latter placement of reward has been regarded as the easier of the two (Moon & Harlow, 1955; Boyd & Warren, 1957; Zentall, Hogan, Edwards, & Hearst, 1980; Bailey & Thomas, 1996).

Rats, thus far, have succeeded in learning oddity using visual stimuli (Wodinsky, & Bitterman, 1953), but have failed to exhibit positive transfer of oddity using odor stimuli (Thomas & Noble, 1988; Bailey & Thomas, 1996). Although, Langworthy and Jennings (1972) claim to have successfully taught rats to learn an oddity problem using odor, odor is very difficult to control and might have provided discriminative cues for the rats. Also, failing to exhibit oddity learning after thousands of training trials are raccoons (Strong & Hedges, 1966). Octopuses (Boal, 1991) were unable to perform at better than chance levels when every trial was novel. Alternatively, pigeons (Zentall, Hogan, Edwards, & Hearst, 1980), gulls and ravens (Benjamini, 1983), a goat (Roitberg & Franz, 2004), and a sea lion (Hille, Dehnhardt, & Mauck, 2004), performed at above-chance levels. Although pigeons were able to perform well on a transfer test with novel stimuli, there was marked impairment in performance from the last training session to the transfer

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trials (Zentall, Hogan, Edwards, & Hearst, 1980). With extensive training, both gulls and ravens were able to successfully perform better than chance on the first trial of a transfer test (Benjamini, 1983). Only one goat of 17 was able to demonstrate transfer on the first trial (Roitberg & Franz, 2004). A sea lion was able to successfully transfer to novel stimuli, however, only after 2,600 training trials (Hille, Dehnhardt, & Mauck, 2004).

Given the mixed results from a variety of studies on oddity learning, an animal's ability to learn oddity may not be the result of a single process, although there always is a possibility that the experimental designs used to test for oddity are somehow flawed. Regardless, there is more evidence for oddity learning in vertebrates than invertebrates. Only one study has been conducted examining oddity learning in an invertebrate, octopus (Boal, 1991). More research needs to be conducted in order to explore whether invertebrates are capable of oddity learning.

The aim of this work is to determine whether honeybees are able to solve an oddity learning task using a simultaneous oddity procedure. Four experiments are described here. Previous pilot work on the oddity problem in this laboratory with honeybees using the traditional simultaneous procedure produced mixed results (Personal communication with P.A. Couvillon: Couvillon, unpublished). The current work differs from the previous work in that new stimuli were used. The new stimuli are composed of two-color compounds in a pinwheel arrangement, or six alternating color wedges, on a Petri dish. These were developed to increase the salience of the stimuli and to better promote attention by the bees. The bees successfully discriminated pairs of the 2 two-colored stimuli in a two-choice procedure, and they did so readily whether the pairs had one color in common or no color in common. It seemed reasonable, then, to use the new stimuli in oddity studies.

In Experiment 1, honeybees were trained in an oddity problem using one odd and two identical nonodd colored Petri dishes. Bees were rewarded with sucrose solution for choosing the odd stimulus. In Experiment 2, honeybees again were trained with the colored Petri dish stimuli and were given an oddity problem using a trial-unique method, different stimuli on each trial. In Experiment 3, honeybees also were trained on an oddity

problem using the trial-unique method of Experiment 2. However, the stimuli of Experiment 3 were displayed on a computer monitor. In Experiment 4, the stimuli were also presented on a computer monitor, but the bees were required to choose the nonodd stimulus.

Experiment 1

Introduction

The aim of Experiment 1 was to determine whether honeybees are able to learn a simultaneous oddity problem using Petri dishes with colored surfaces. The plan was to first try to demonstrate oddity learning and then to analyze it in subsequent experiments. In each trial, bees were presented with two nonodd stimuli and one odd stimulus. In order to receive a sucrose reward, the bees had to choose the stimulus that was different from the other two. Alternatively, stevia, which is an aversive substance, was placed on the two nonodd stimuli in each trial. This experiment included two groups of bees; one group was trained with a pair of two-color compounds with a common color, and the other group with a pair of two-color compounds with no common color. It seemed reasonable that the discrimination of oddity might be easier with no color in common.

Subjects

The subjects were 24 honeybees (*Apis mellifera*) never used in prior experiments. They were captured at feeders containing 10-20% sucrose solution near the hives in back of the Békésy Laboratory at the University of Hawai'i at Mānoa. The subjects were assigned to two groups, No Color in Common group (NCC) and One Color in Common group (OCC). The bees in each group were trained individually in a single daily session lasting from one to several hours.
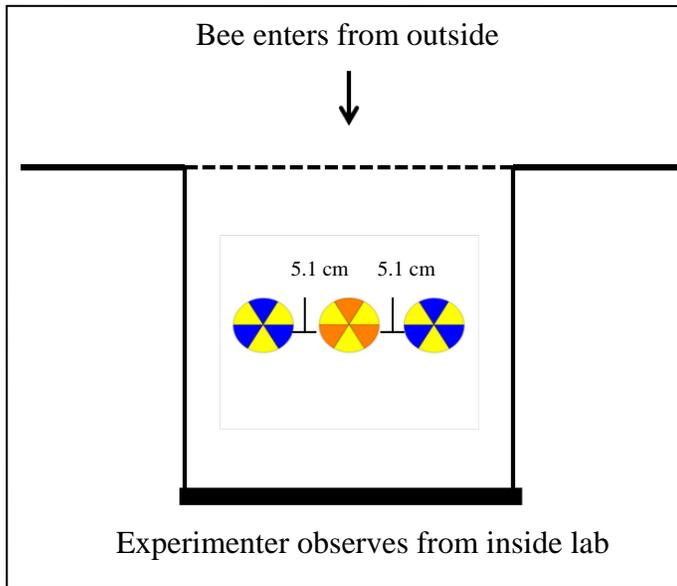
Figure 1. Apparatus used for Experiments 1 and 2.

The main apparatus used for training, shown in Figure 1, was a wooden enclosure, 61 cm wide, 61 cm high, and 61 cm deep, recessed in the exterior wall of a laboratory window. Two sliding Plexiglas partitions separated the interior of the laboratory from the exterior, which prevented both unwanted entrance of the bees into the room and permitted observation by the experimenter.

The stimuli used were Petri dishes 5.5 cm in diameter. Patterns of six equally segmented triangles made of vinyl plastic with a matte-finish were placed on each dish using silicon sealant. Dishes displayed two of the following colors in alternating sequence: blue (B), green (G), orange (O), and yellow (Y). All four colors had been shown in previous experiments conducted in this laboratory to be highly discriminable and equally preferred. In addition, previous color discrimination experiments conducted in the laboratory have shown that each combination of two-color compound stimuli can be discriminated. There are six two-color compound stimuli and fifteen total pairs of two-

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colored compounds. Due to the nature of the fifteen combinations of two-color compound stimuli, three combinations have no color in common and twelve have one color in common. For this reason, two groups of bees were assessed, group No Color in Common (NCC) and group One Color in Common (OCC). In each trial, three stimuli, two identical and one different, were presented simultaneously, positioned in the center of the floor of the wooden apparatus, 5.1 cm apart edge-to-edge parallel to the window edge, as shown in Figure 1.

Procedure

Pretraining: A forager bee that was collected in a matchbox from the feeder was brought into the laboratory and released on a pretraining stimulus (one of the two stimuli to be used in training) in the middle of the floor of the wooden enclosure. A 100-µl drop of 50% sucrose was placed on top of the pretraining stimulus. As the bee was feeding it was marked on the thorax with colored enamel for identification purposes. The honeybee drank until replete and then flew to the hive (usually returning in 3 to 5 minutes). A timer was used to record the roundtrip return time of the experimental bee. If the honeybee returned to the window, it found another drop of 50% sucrose on the second pretraining stimulus (the other stimulus to be used in training). If it did not return, either it was captured again from the feeding station, or another bee was collected. This process was repeated until a bee reliably returned to the pretraining stimuli.

Training: After the pretraining phase, training began. In each training trial, the bee found three stimuli, two identical and one different. The two identical, nonodd stimuli always had a100-µl drop of 10% stevia solution placed centrally on top of the stimulus. Previous experiments conducted in this laboratory provided evidence that stevia is aversive to bees. For this reason, stevia was used in training to "punish" bees for choosing an incorrect stimulus and to ensure that each stimulus had a drop placed on top for perceptual similarity. (Pilot work in this laboratory showed that honeybees cannot

discriminate a stevia drop from a sucrose drop on a Petri dish without tasting.) The one different, odd stimulus always had a 100-μl drop of 50% sucrose solution placed on top. Sucrose was used on the odd stimulus to reinforce the bees for choosing the correct stimulus.

Only a bee's initial choice in each trial was recorded and used for analysis. Correct responses were those where the bee landed and proceeded to drink the sucrose on the odd stimulus. If the bee chose correctly, it flew back to the hive, regurgitated its meal, and returned for another trial. A correction procedure was used here; if the bee chose incorrectly, it was allowed to fly (or in rare circumstances walk) to its next choice until the correct choice was made. If the bee touched or drank the stevia from either of the nonodd stimuli, an error was recorded.

Individual bees in both groups received a single session that consisted of 15 trials, and each session was balanced for position. Six possible spatial arrangements of stimuli (e.g., ABB, ABA, BAB, AAB, BBA, BAA) were used to prevent position discrimination and to promote oddity discrimination. Reward occurred five times in each position (left, middle, right) in a quasi-random sequence over the 15 training trials. For each bee, on seven of the trials one of the two-color compounds was odd, and on the other eight trials the other two-color compound was odd, with trials of the two kinds intermixed in a quasi-random sequence. Identical two-color compounds could be rewarded on two trials in a row but only in different positions.

Results & Discussion

The performance of the two groups is plotted in Figure 2 in terms of the proportion of bees in each group that chose correctly across training trials; the line at .33 indicates chance. The trial-by-trial results for group NCC are shown in the left panel and for group OCC in the right panel. Both groups show better than chance performance. The mean proportion correct choices over 15 trials for group NCC was .42 and was significantly greater than chance [$t(11) = 2.28$, $p < .05$]. The mean proportion correct

choices over 15 trials for group OCC was .50 and was significantly greater than chance [t(11) = 3.90, p < .01].
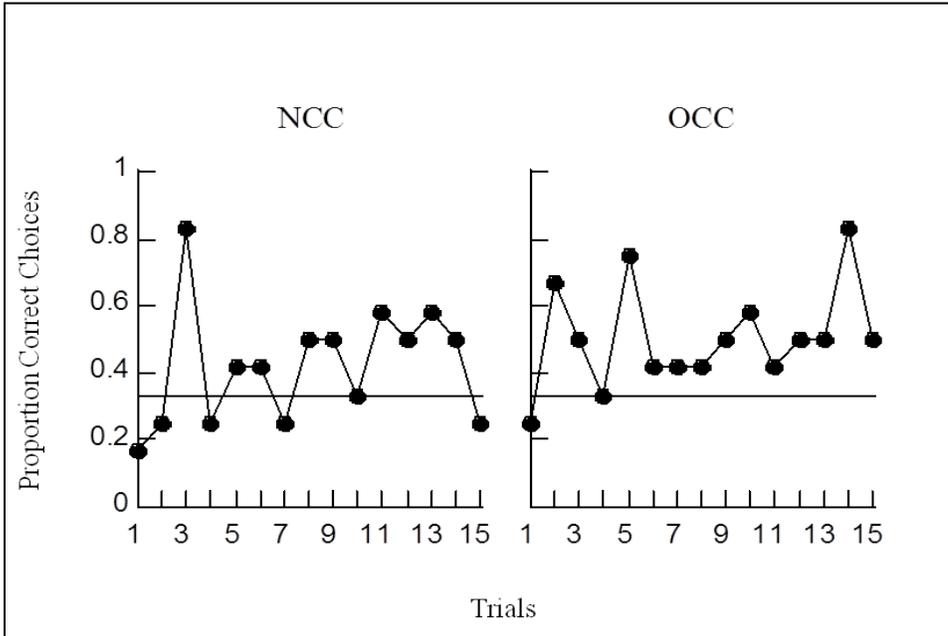

Figure 2. Results for the two groups trained in Experiment 1.

Although performance in the OCC group appears to be better than that of the NCC group, there was no significant difference in the overall proportion correct choices between the two groups [t(22) = -1.40, p > .05]. These results suggest that honeybees treat the two-color stimuli as compounds. The results do suggest that honeybees are responding on the basis of oddity. However, the performance of both groups also suggests that the discrimination is difficult.

It is common in vertebrate studies of choice discrimination learning in difficult problems to further analyze the results to determine if the subjects used systematic strategies (Moon & Harlow, 1955). Therefore it seemed prudent to further analyze the bees' choices to determine first if there was a significant position or color preference. It

also seemed prudent to look for any tendency on a given trial to choose the rewarded position or the rewarded stimulus of the preceding trial.

Position preference, the tendency to choose one position (left, middle, right) more than another position, was analyzed. If bees do not have a position preference, it is expected that they will have equal frequencies of initial position choices across all training trials. However, a chi-square test for equal frequencies showed significant preference in both groups [NCC, $\chi^2 (2) = 7.59$, p < .05; OCC, $\chi^2 (2) = 12.69$, p < .005]. In group NCC, there was a tendency to choose the middle position, and, in group OCC, a tendency to choose the left position.

Stimulus preference, that is, the tendency to prefer one color compound over the other, was also analyzed. It is expected that bees with no stimulus preference will choose each two-color compound 50% of the time. Analysis of the bees' stimulus preference using a one-sample t-test yielded mixed results. Overall, group NCC did not have a stimulus preference [t(11) = .522, p >.05], although 5 out of 12 bees clearly demonstrated a preference indicated by 5 more choices to one stimulus over another. On the other hand, group OCC overall had a significant stimulus preference [t(11) = 2.45, p < .05], although only 2 out of 12 bees had a clear preference. These results, taken together, suggest that some of the individual bees in each group had a stimulus preference, that is, a nonrandom choice of stimulus on any given trial.

Next, the tendency of position reward following was analyzed. Position reward following is when a subject chooses the same position on trial n+1 that had been rewarded on trial n. For example, if on trial n the left position was rewarded, position reward following on trial n+1 would be indicated by choice of the left position. To determine whether either group of bees had a tendency across trials for position reward following, the bees' proportion of initial choices following a previously rewarded position was analyzed; chance should be .33. The mean proportion for NCC was .30 and was not significantly different than chance [t(11) = -1.05, p > .05]. The mean proportion for group OCC was .28 and was not significantly different than chance [t(11) = -1.34, p > .05]. Neither group chose a position based on immediate prior reward of that position.

Finally, stimulus reward following was analyzed. Stimulus reward following was analyzed exactly the same way as position reward following. For example, if on trial n stimulus A was rewarded, stimulus reward following on trial n+1 would be indicated by choice of stimulus A. To assess the possibility that the honeybees could be making responses based on stimulus reward following, the bees' proportion of initial choices of the previously rewarded stimulus was analyzed; chance should be .50. Analysis of both groups did not yield significant results. The mean proportion for group NCC was .56 and was not significantly different than chance [$t(11) = 1.61$, $p > .05$]. The mean proportion for group OCC was .43 and was not significantly different than chance [$t(11) = -2.05$, $p > .05$]. Neither group chose a stimulus based on immediate prior reward of that stimulus.

In summary, both groups show a small, but significant tendency to choose the odd stimulus across the 15 training trials. Position preferences were found in both groups. Stimulus preferences, however, were only shown in group OCC and not in group NCC. Analysis of position and stimulus reward following did not yield any significant results.

The small tendency to choose the odd stimulus could be due to conditional discrimination learning or it could be due to oddity learning. Conditional discrimination learning needs to be ruled out if the results are to be interpreted as evidence for oddity learning. The oddity problem presented in this experiment can be learned based on conditional discrimination; if BB choose A and if AA choose B. Traditionally, as in matching-to-sample (MTS) procedures, a nonrewarded transfer test is needed to demonstrate concept learning and to rule out conditional discrimination learning. To rule out conditional discrimination learning in an oddity problem, a nonrewarded transfer test to novel stimuli can be used. For example, if the bees were trained on A+ B- B- (reward indicated by +) and B+ A- A-, then transfer could be to C+ D- D-. On the transfer test, choice of stimulus C would indicate successful transfer. A transfer test may not be feasible here because the performance in training was so variable. Another commonly recognized method for testing for concept learning is a trial-unique procedure (Wright, Cook, Rivera, Sands, & Delius, 1988), where every trial is different. It should be noted that there is almost no evidence in the vertebrate literature for successful trial-unique

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performance in any discrimination including oddity and MTS. However, work on MTS and NMTS with bees (Shishimi, doctoral dissertation, in progress) found successful MTS and NMTS with a trial-unique procedure. On that basis, it seemed reasonable to try an oddity problem with a 'trial-unique' procedure, where every trial is a transfer test, rather than attempt a replication of Experiment 1 followed by a transfer test with novel stimuli.

Experiment 2

Introduction

The results of Experiment 1 suggest a weak oddity learning effect, however, it is difficult to interpret. The aim of this experiment was to try a trial-unique procedure, an alternative to a transfer test, using the same kind of stimuli as used in Experiment 1. Every trial consists of a different combination of stimuli (e.g., ABB, DCC, EAA, CFF, BEE, FDD, EBB, DBB, FAA, EDD, CBB, FEE, ADD, BFF, CAA) such that in each trial, bees were presented with two nonodd stimuli and one odd stimulus. Better than chance choice performance would provide strong evidence of oddity learning.

Subjects

The subjects were 12 honeybees (*Apis mellifera*) never used in prior experiments. They were captured at feeders containing 10-20% sucrose solution near the hives in back of Békésy Laboratory at the University of Hawai'i at Mānoa. Each subject was trained individually in a single daily session lasting from one to several hours.

Apparatus & Stimuli

The same apparatus and stimuli used for training in Experiment 1 (see Figure 1) were used here. Two two-color compound arrangements of four possible colors produce 15 unique combinations allowing for each trial in a 15-trial session to be unique. As Wright, Cook, Rivera, Sands, & Delius (1988) note, a trial-unique procedure may enhance the speed of learning acquisition by providing numerous training exemplars.

All bees received a single session that consisted of 15 training trials. There were four different trial sequences of the stimulus configurations, each used for three subjects. The sequences were constructed so that successive trials did not share any identical

stimuli, ensuring that the same two-color compound was not rewarded twice in a row (see Table 1 for a sample trial sequence). Reinforced two-color compounds were never presented in the same position more than once in the 15-trials. In each trial, three stimuli -- two identical and one different -- were presented simultaneously, positioned in the center of the floor of the wooden apparatus, 2.55 cm apart edge-to-edge parallel to the window edge. (Note that to reduce the likelihood of position biases in the bees' choices, the distance between the stimuli was decreased from the 5.1 cm used in Experiment 1.)

Procedure

Pretraining: Pretraining was exactly the same as in Experiment 1, except the pretraining stimulus consisted of four equal segments of each color to be used in training (blue, yellow, orange, and green).

Training: Training was the same as in Experiment 1, except that different pairs of two-color stimulus compounds were presented on each training trial (see Table 1).

Results & Discussion

The performance of the bees is plotted in Figure 3 in terms of the proportion of bees that chose correctly across training trials; the line at .33 indicates chance. The mean proportion correct choices over 15 trials was .49, which is significantly greater than chance [$t(11) = 4.55$, $p = .001$]. Taken at face value, this result is evidence for oddity learning in honeybees. Nonetheless, given the position preference of the bees in Experiment 1, it was necessary to further analyze the bees' choices to determine if there was a significant position preference or a tendency for position reward following.
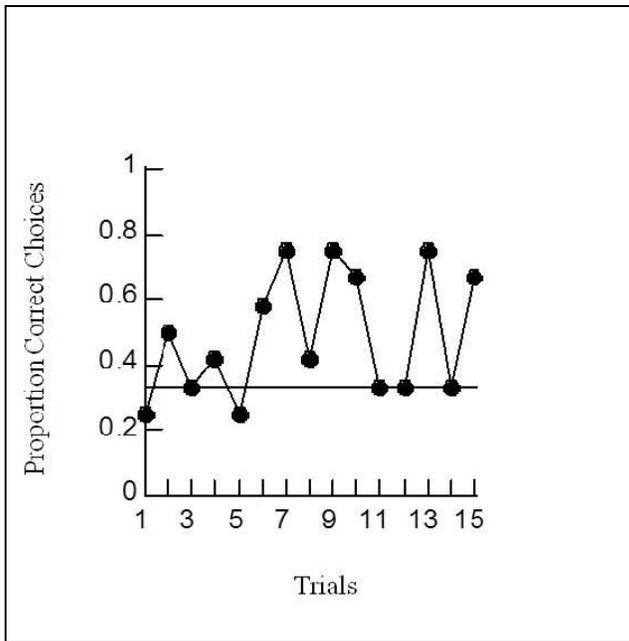
Figure 3. Results of Experiment 2.

Position preference, the tendency to choose one position (left, middle, right) more than another position, was analyzed. If bees do not have a position preference, it is expected that they will have equal frequencies of initial position choices across all training trials. A chi-square test for equal frequencies showed no significant position preference [$\chi^2(2) = 2.50$, $p > .05$], suggesting that the bees have no systematic tendency to choose any one of the three positions.

Position reward following also was analyzed to determine whether the bees had a tendency on any given trial to follow the position rewarded on the preceding trial; chance was set at .33. The mean proportion of position reward following was .25 which was significantly less than chance [$t(11) = -3.07$, $p < .01$]. This result suggests that the bees have a strong tendency to <u>not</u> follow a previously rewarded position. Instead of choosing the same position, they are choosing a different position, that is, they are switching. It is

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not clear if the bees' position-switching is due to active avoidance of the previously rewarded position. Since the trial sequences were constructed (see Table 1) such that the position of the rewarded stimulus was rarely the same on two successive trials, the bees may simply have learned not to revisit the rewarded position of the previous trial.

In conclusion, the results of this experiment provide strong evidence of oddity learning in honeybees. Analysis of position preference did not yield significant results, although there was a significant tendency to not choose the previously rewarded position. It is not possible to determine if the 'switching' behavior exhibited by the bees is due to a nonrandom choice tendency or is an artifact of learning to choose on the basis of oddity. The oddity learning found in this experiment cannot be explained by associative learning. Since every trial is unique, bees are not able to learn the oddity problem based on conditional discrimination, although it is possible that the bees have a preference for odd or "different" stimuli. Therefore, it is important also to demonstrate that honeybees can solve a nonoddity problem, that is, reward for choice of one of the two nonodd stimuli.

In the subsequent experiments reported here, a computer monitor with PowerPoint generated images is used to display the stimuli. The new stimulus display method provides more options for generating stimuli and for manipulating their presentation. Experiment 3 is a replication of Experiment 2 with the computer-generated stimuli. If the replication proved to be successful, the plan was to then conduct a nonoddity experiment with the same computer-generated stimuli.

Experiment 3

Introduction

The aim of this experiment was to replicate Experiment 2 using a computer monitor to generate stimuli, which allows for easier manipulation and more options for spatial presentation of stimuli, as well as for varying the number of stimuli. This experiment used the same trial-unique procedure that was used in Experiment 2. In each trial, bees were presented with two nonodd stimuli and one odd stimulus.

Subjects

The subjects were 12 honeybees (*Apis mellifera*) never used in prior experiments. They were captured at feeders containing 10-20% sucrose solution near the hives in back of Békésy Laboratory at the University of Hawaiʻi at Mānoa. Each subject was trained individually in a single daily session lasting from one to several hours.
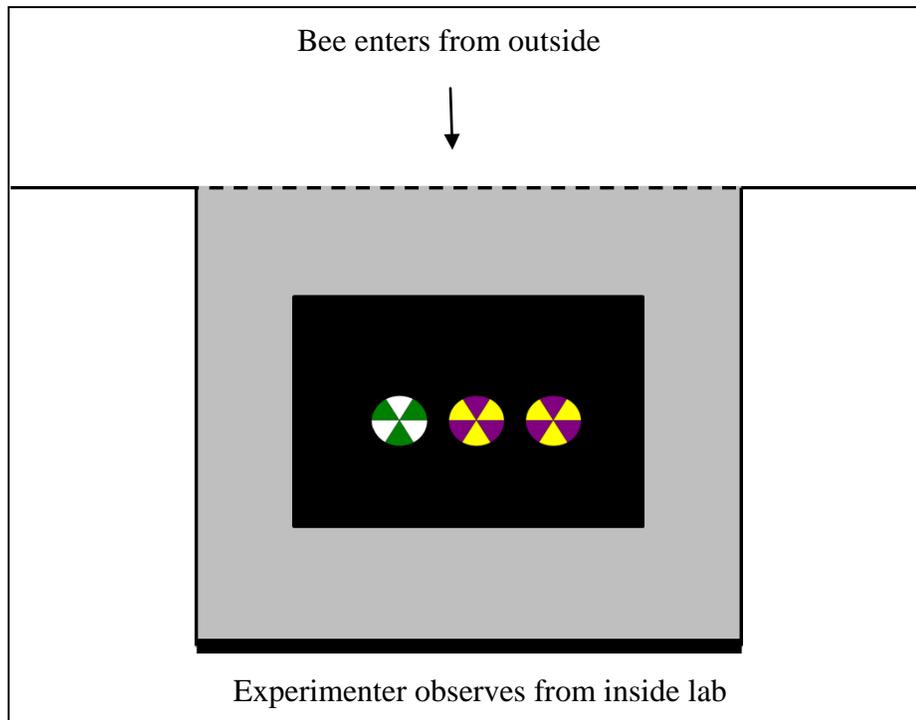
Figure 4. Apparatus used for Experiments 3 and 4.

The main apparatus used for training, shown in Figure 4, was a wooden enclosure, 61 cm wide, 61 cm high, and 61 cm deep, recessed in the exterior wall of a laboratory window. A flat screen 15-inch computer monitor with a glass surface replaced the floor of the apparatus. The monitor was covered by a thin piece of plywood (painted gray) with an opening cut out to expose the display. The visible display surface was recessed into the plywood covering 1.5 cm. The measurements of the visible display surface were 21.6 cm by 29.2 cm. Two sliding Plexiglas partitions separated the interior of the laboratory from the exterior, which both prevented unwanted entrance of the bees into the room and permitted observation by the experimenter.

The shape, size, and distance of the two-color compound stimuli used in Experiment 2 were approximated with a series of slides generated in PowerPoint. The

displayed stimuli were 6.5 cm by 5 cm in diameter and were positioned 2 cm apart edge-to-edge parallel to the window edge, as shown in Figure 4. Patterns of six equally segmented wedges were displayed on each stimulus. Stimuli displayed two of the following colors in alternating sequence: Purple (P), green (G), yellow (Y), and white (W), all of which had been used in previous discrimination experiments conducted in this laboratory using computer-generated images and shown discriminable. The colors were created in Microsoft Paint by specifying values for yellow, white, green, and purple. The values are as follows: **yellow** (red = 255, green = 255, blue = 0; hue = 40, saturation = 240, luminosity = 120), **white** (red = 255, green = 255, blue = 255; hue = 160, saturation = 0, luminosity = 240), **green** (red = 0, green = 153, blue = 0; hue = 80, saturation = 240, luminosity = 60), **purple** (red = 153, green = 0, blue = 153; hue = 200, saturation = 240, luminosity = 72). To facilitate the bees' detection of the stimuli, the background of the PowerPoint slides was black to maximize contrast with the colored stimuli.

Procedure

Pretraining: Pretraining was the same as in Experiment 2, that is, exposure to a stimulus compound of the four colors to be used in training. The pretraining stimulus was displayed as equal segments of yellow, white, green, and purple.

Training: Training was the same as in Experiment 2, except that a 15% salt solution was used on the nonrewarded stimuli instead of the stevia solution used before. Prior experiments in this laboratory indicated that a drop of stevia can be discriminated from a drop of sucrose on a computer monitor, but a drop of salt solution cannot be discriminated from a drop of sucrose. Also different from Experiments 1 and 2 were the recording measurements used to indicate correct and incorrect choice. Since the bees often walked on the computer monitor, a correct choice was measured as drinking from the odd stimulus. An incorrect choice was measured as drinking from either of the nonodd stimuli. (See Table 1 for a sample trial-unique sequence.)

Results & Discussion

The performance of the bees is plotted in Figure 5 in terms of the proportion of bees that chose correctly across training trials; the line at .33 indicates chance. The mean proportion correct choices over 15 trials was .51, which is significantly greater than chance [$t(11) = 5.05$, $p < .000$]. This result replicates that of Experiment 2 and provides additional evidence for oddity learning with trial-unique stimuli. As in Experiment 2, the bees' choices were further analyzed to determine if there was a significant position preference or a tendency for position reward following.
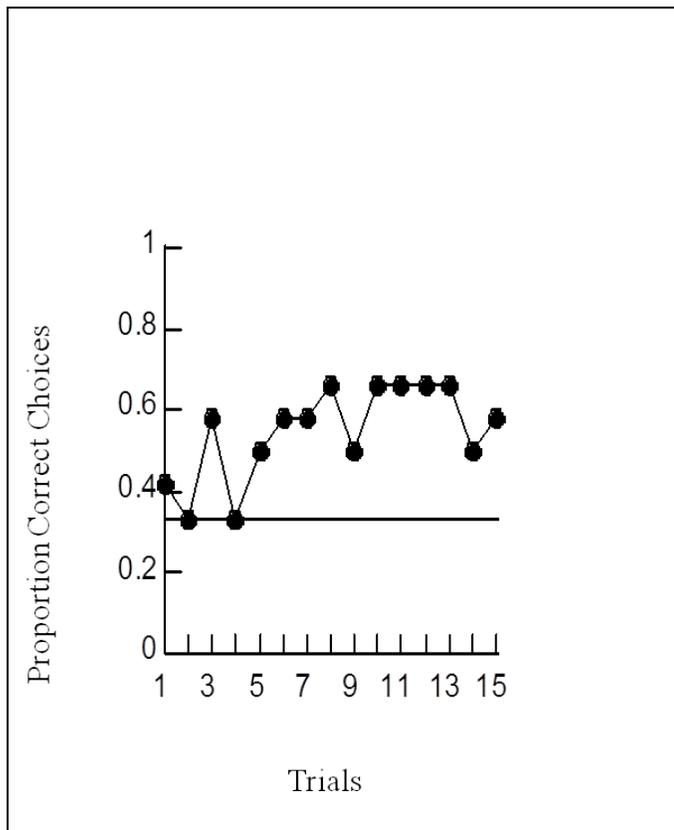

Figure 5. Results for Experiment 3.

Position preference, the tendency to choose one position (left, middle, right) more than another position, was analyzed. If bees do not have a position preference, it is expected that they will have equal frequencies of initial position choices across all training trials. A chi-square test for equal frequencies showed a significant position preference [$\chi^2(2) = 9.72$, $p < .01$], suggesting that the bees have a systematic tendency to choose left position.

Position reward following also was analyzed to determine whether the bees had a tendency on any given trial to follow the position rewarded on the preceding trial; chance was set at .33. The mean proportion of position reward following was .31 which was not significantly different than chance [$t(11) = -.519$, $p > .05$]. The bees did not chose a position based on immediate prior reward of that position.

In conclusion, the results of this experiment again provide evidence of oddity learning in honeybees, here with computer-generated stimuli. In the next experiment honeybees were trained in a nonoddity problem, where choice of either of the two identical stimuli was rewarded and choice of the odd stimulus was not rewarded. If the oddity learning seen in the previous experiments was enhanced by an unlearned preference for oddity, then honeybees should not be able to learn the nonoddity problem, or, at least, learning should appear to be more difficult than in the oddity problem.

Experiment 4

Introduction

The aim of this experiment was to determine whether honeybees could learn to choose on the basis of nonoddity. This experiment used the same trial-unique procedure as used in Experiment 3 except a reward was provided on both of the nonodd stimuli and salt solution was placed on the odd stimulus. The results of this experiment are important for an understanding of the oddity learning demonstrated in Experiments 2 and 3. If oddity learning is based mostly on an unlearned preference for oddity in honeybees, then the nonoddity problem should be difficult, if not impossible. However, if the bees also solve the nonoddity problem, the results for both oddity and nonoddity may be based on concept learning, which, as noted above, refers to learning that is not readily accountable for by associative learning principles.

Subjects

The subjects were 12 honeybees (*Apis mellifera*) never used in prior experiments. They were captured at feeders containing 10-20 % sucrose solution near the hives in back of Békésy Laboratory at the University of Hawaiʻi at Mānoa. Each subject was trained individually in a single daily session lasting from one to several hours.

Apparatus & Stimuli

The same apparatus used for training in Experiment 3 was used here, and the stimuli also were the same.

Procedure

Pretraining: Pretraining was the same as in Experiment 3.

Training: Training was the same as the trial-unique training of Experiment 3, except 50% sucrose reward was provided on the two nonodd stimuli and an aversive 15% salt solution on the odd stimulus. (See Table 1 for a sample trial-unique sequence.)

Results & Discussion

The performance of the bees is plotted in Figure 6 in terms of the proportion of bees that chose correctly across training trials; the line at .66 indicates chance. Chance is now .66 because the bees are given two rewarded stimulus compounds. The mean proportion correct choices over 15 trials was .75, which is significantly greater than chance [$t(11) = 2.88$, $p < .05$]. This result clearly indicates that honeybees can learn a nonoddity problem. As in the previous experiments, the bees' choices were further analyzed to determine if there was a significant position preference or a tendency for position reward following.
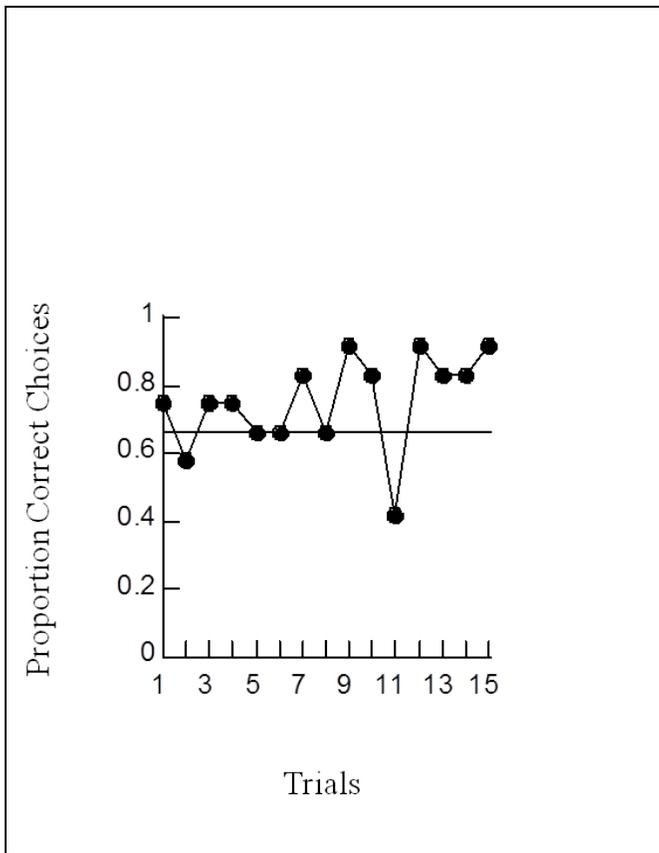
Figure 6. Results for Experiment 4.

Position preference, the tendency to choose one position (left, middle, right) more than another position, was analyzed. If bees do not have a position preference, it is expected that they will have equal frequencies of initial position choices across all training trials. A chi-square test for equal frequencies did not show a significant position preference [$\chi^2(2) = 1.58$, p > .05], suggesting that the bees do not have a systematic tendency to choose any one of the three positions.

Position reward following also was analyzed to determine whether the bees had a tendency on any given trial to follow the position rewarded on the preceding trial; chance was set at .33. The mean proportion of position reward following was .31 which was not significantly different than chance [$t(11) = -.56$, p > .05]. The bees did not chose a position based on immediate prior reward of that position.

In conclusion, the results of this experiment provide evidence of nonoddity learning in honeybees. It is worth noting that the likelihood of choosing correctly by chance is very high, .66. Even in simple choice discrimination problems honeybees rarely choose correctly 100% of the time, often achieving about .90 proportion of correct choice. It is noteworthy that the bees in this experiment performed significantly better than chance (.75) given the possibility of a ceiling effect. The fact that the bees readily solved the nonoddity problem argues against the possibility that the bees have an unlearned preference for oddity. That honeybees show both oddity and nonoddity learning with trial-unique stimuli is a compelling case for concept learning.

General Discussion

The purpose of the four experiments reported here was to determine if honeybees could learn an oddity problem. Robinson (1933) first established that a simultaneous oddity problem was one way to study concept learning in nonhuman animals. These experiments are the first serious attempt to examine whether honeybees can learn oddity.

Experiment 1 was a variant of Robinson's (1933) classical 2-stimulus oddity problem, where reward occurs on stimulus A on one kind of trial (A + B- B-) and on stimulus B on the other kind of trial (B+ A- A-). Honeybees given this problem were trained with colored Petri dishes and rewarded with sucrose for choice of the odd color on each trial. The results showed that honeybees were able to perform at levels greater than chance. However, their small tendency to choose the odd stimulus could have been due to conditional discrimination learning rather than to oddity learning.

Experiment 2 was conducted in order to rule out conditional discrimination learning. Usually to rule out conditional discrimination, transfer trials with new stimuli (e.g., C+ D- D- and D+ C- C-) are given to experimental subjects after training with A+ B- B- and B+ A- A- . Correct choice on the first transfer trial is typically taken as evidence for oddity or concept learning. Another method is to use unique stimuli on every trial. The trial-unique method is regarded, in the vertebrate literature, as the "gold standard" to test for concept learning. Unpublished work in this laboratory (Shishimi, doctoral dissertation, in progress) found successful trial-unique matching-to-sample and non-matching-to-sample performance in honeybees. Therefore, it seemed reasonable to use this approach with an oddity problem. The trial-unique method was used in Experiment 2, and honeybees were able to perform at levels greater than chance. These results cannot be explained as conditional discrimination learning and provide strong evidence of oddity learning in honeybees.

Experiment 3 was a replication of Experiment 2 using the trial-unique stimulus method with new stimuli displayed on a computer screen. The honeybees again were able to successfully learn the oddity problem at levels greater than chance. These results,

which replicate those of Experiment 2, again cannot be explained as simple conditional discrimination because the stimuli change on every trial. The only other possible explanation of the honeybees' greater than chance performance is that honeybees could have an unlearned preference for odd or "different" items. The issue of whether oddity learning could be explained by such a preference still needed to be addressed.

Experiment 4 was designed to determine if honeybees could solve a nonoddity problem. If the oddity learning found in Experiments 2 and 3 was based only on a preference for choosing the odd stimulus, then a nonoddity problem should be difficult for honeybees. The same procedure and trial configurations of Experiment 3 were used in Experiment 4, but reward occurred on the nonodd stimuli instead of the odd stimulus. The greater than chance performance indicates that honeybees are able to learn a nonoddity problem.

In summary, the experiments reported here provide compelling evidence of both oddity and nonoddity learning in honeybees. These results, taken together, cannot be explained by associative learning (conditional discrimination) or unlearned preference for novelty. The results do suggest that honeybees may have learned to discriminate oddity and nonoddity on the basis of concepts.

Traditionally, learning research conducted with invertebrates has been primarily concerned with exploring basic learning phenomena using analogs of vertebrate learning experiments. This approach was used here, that is, to explore how honeybees perform in oddity problems that are analogous to those used with vertebrates. The honeybees' successful performance here is noteworthy because vertebrate performance in oddity problems is highly variable, sometimes successful and sometimes not (for example see, Boyd & Warren, 1957; Warren, 1960; Strong & Hedges, 1966). Comparison of experimental details between the honeybee experiments and those of the vertebrate experiments point to two methodological differences that may contribute to the differences in performance seen across species on oddity problems.

One methodological difference is that vertebrate oddity experiments typically have massed trials, whereas, the above experiments with honeybees have spaced trials.

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Oddity training with pigeons, for example, is conducted over many daily sessions with 20 to 30 trials per session. The intertrial interval typically ranges from 10 to 30 seconds. The oddity training with honeybees was conducted in a single session with 15 trials. The intertrial interval averaged 3 to 5 minutes. (Note that the intertrial interval for honeybees was determined by the time it took for the bee to leave the window, deposit the sucrose at the hive, and return to the window for another trial.) Highly massed trials can lead to both proactive and retroactive interference that may contribute to the variable performance seen across the different vertebrate species. Spaced trials may minimize proactive and retroactive interference, and, therefore, contribute to honeybees' successful performance on the oddity and nonoddity problems. Further research needs to be conducted to determine the extent to which massed and spaced trials affect performance in oddity problems.

Another methodological difference between the vertebrate and honeybee experiments is the use of punishment. Typically in vertebrate experiments, subjects are given reward for choosing a correct stimulus and no reward for choosing an incorrect stimulus. In the honeybee experiments, subjects were given reward (sucrose) for choosing a correct stimulus but also were punished with stevia or salt for choosing an incorrect stimulus. The use of punishment may facilitate learning because it may increase the cost of incorrect choice which also could increase attention to the stimuli.

Since the goal of comparative research with honeybees is to compare their performance to that of vertebrates in similar problems, it is useful to review the theoretical interpretations that have been discussed in the vertebrate literature. Concept learning has been mentioned here as a possible basis for the successful performance of the oddity and nonoddity problems by honeybees. However, "concept" is only one term that is used in the vertebrate literature, but is by no means standard. Other terms in the literature include, relational discrimination, stimulus relations, relational concepts, events and their interrelations, abstract concepts, learning relations about relations, higher-order relations, and generalized concepts (Cook and Wasserman, 2012). The conflicting definitions and terminology for "concept" may not just be a difference of style or

phrasing. For example, both Cook and Wasserman (2012) have conducted experiments on concept learning in pigeons. Both experimenters use Same/Different (S/D) discriminations in their respective laboratories. However, Cook (2012) uses a multi-stimulus discrimination array to test for concepts, whereas, Wasserman (2012) uses an icon-based discrimination array to test for concepts. Both Cook and Wasserman (2012) use the same term "concept" to explain the successful performance on the S/D discrimination problem by their pigeons. However, Wasserman (2012) considers another possibility, entropy, "an information theoretic concept," defined operationally as "item variability." Wasserman (2012) has empirical evidence that an increase in the number of icons in his display (increase in entropy) improves performance in S/D problems. Cook (2012), however, fails to find such improvement, and, in fact, finds deterioration in performance with an increase in the number of stimuli in his arrays. Cook and Wasserman (2012) conclude, "…different tasks rely on different perceptual and conceptual processes." Whether honeybees are using perceptual or conceptual processes still needs further examination. Given the similarities in display types between the stimuli used here and Cook's (2012) stimuli, it is possible that honeybees may solve the oddity and nonoddity problem using concepts.

Table 1. Sample trial sequence for Experiment 2.
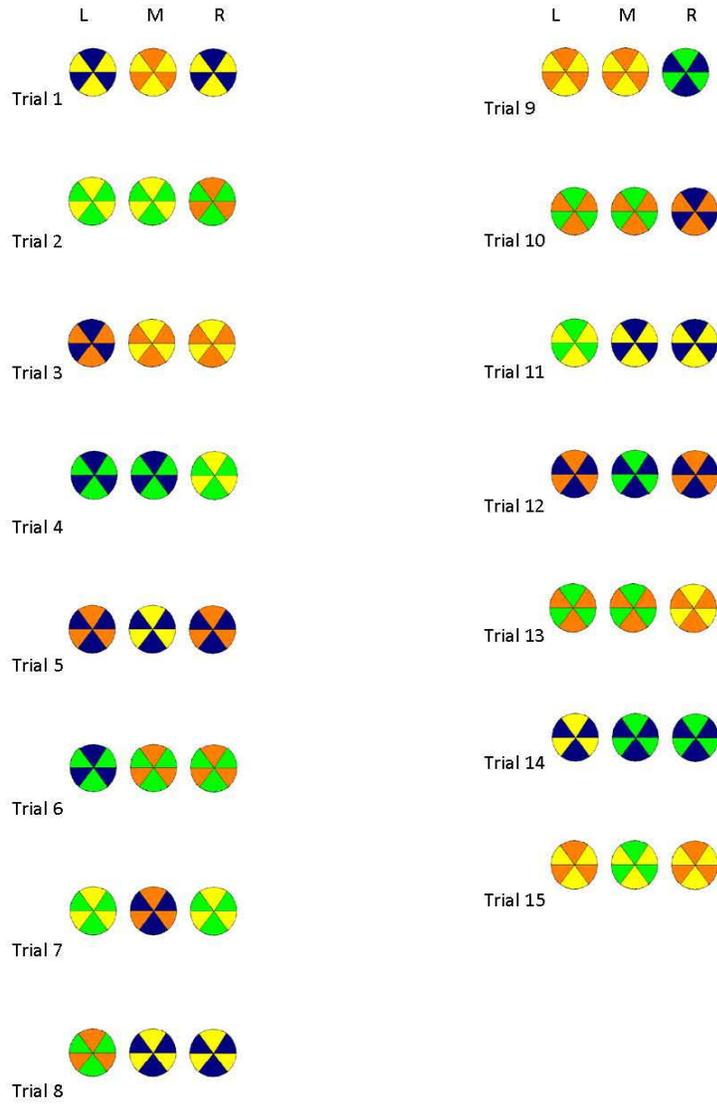
L (left), M (middle), R (right) refer to stimulus position in the experimental window.

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