Reward expectations in honeybees

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The study of expectations of reward helps to understand rules controlling goal-directed behavior as well as decision making and planning. I shall review a series of recent studies focusing on how the food gathering behavior of honeybees depends upon reward expectations. These studies document that free-flying honeybees develop long-term expectations of reward and use them to regulate their investment of energy/time during foraging. Also, they present a laboratory procedure suitable for analysis of neural substrates of reward expectations in the honeybee brain. I discuss these findings in the context of individual and collective foraging, on the one hand, and neurobiology of learning and memory of reward.

Expectations of Reward

An expectation is said to be “a strong hope or belief that something that you want will happen”, or the action “to anticipate or look forward to the occurrence of an event.” Clearly, the notions of expectation and anticipation are linked to each other and it is taken for granted that these two words are interchangeable. However, although one needs to expect in order to anticipate, expecting per se does not imply anticipation. In psychology, the term ‘incentive’ designates internal correlates of specific rewards guiding the behavior of subjects pursuing such rewards. Incentive terms closely mimicking a natural situation. Using an approach frequent in behavioral ecology, they helped to ponder the role of reward expectations in the ecology of foraging. The remaining two studies concern a laboratory procedure suitable for analysis of neural substrates of reward expectations in the honeybee brain. They made use of an approach frequent in the study of reward expectations, which relies heavily on experiments with restrained subjects made under highly controlled conditions. Using this approach, for example, studies in mammals have shown that interaction between the basolateral complex of the amygdala and the orbitofrontal cortex is necessary for development and subsequent use of reward expectations involved in goal-directed behaviors. I shall discuss these studies in the context of individual and collective foraging, on the one hand, and the neurobiology of learning and memory of reward.

Reward Expectations in Foraging Honeybees

Honeybees live in large colonies whose primary source of energy is the nectar found within flowers. Nectar offers vary continuously, both in space and time, depending on species-specific flowering patterns, weather conditions, and the activity of other pollinators. In spite of such variability, honeybees gather energy efficiently using their learning and memory skills. They learn, for example, the location and time of day when flowers are productive as well as their odors, colors and shapes. But, can they also learn that reward level increases or decreases over time? In a recent study, we trained bees to forage individually on an artificial flower patch offering increasing (small-medium-large), decreasing (large-medium-small) or constant (small, medium or large) reward levels (Fig. 1A). Next, after a long foraging pause, we recorded how consistently they searched for food at the patch in the absence of reward. We found that...
Honeybees, therefore, adjust their persistence to search for food in relation to both the sign and magnitude of past variations in reward level. The outcome of the above experiments is shown schematically in Figure 2. When honeybees forage on a flower patch offering variable reward levels, two parallel learning processes take place. On the one hand, bees learn the sign and magnitude of reward variations across successive foraging visits. They may do this using a build-in change detector that computes the difference in reward magnitude across foraging events. This computation leads to an estimate of an expected reward; we refer to such estimate as to a reward memory. On the other hand, bees associate the offered reward (as the US) with signals and cues present at the feeding site (as CSs) and an associative memory is formed. When a bee visits the feeding site after a long foraging pause, these memories are retrieved by reward-predicting stimuli. Associative memories are revealed through the bee's choice behaviour, whereas reward memories are revealed through the bee's persistence to search for food in the absence of reward. Hence, foraging honeybees adjust their investment of time/energy during food searches in relation to both the sign and magnitude of past variations in reward level.

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phenomena into the context of foraging. One such model incorporates reward level variability into the forager's evaluation of food patch quality.35 It predicts that the foraging behavior of animals that have previously experienced variable rewards at a given patch will depend upon their memories of either the most recent reward level or the average reward level experienced, depending on the time elapsed since the last encounter with reward.35 Our results do not match predictions from this model.12,13 Therefore, an alternative model is needed to explain how honeybees use reward memories during foraging.

Reward Expectations in Harnessed Bees

In addressing neural correlates of reward expectations in honeybees, one has to find a behavioral correlate of reward memories suitable for laboratory studies. Such behavioral correlate can be the honeybee proboscis extension response (PER).36-38 This response allows bees to gather sugar solution and is triggered when the gustatory receptors of the antennae, proboscis and tarsi are stimulated with sucrose.37 In a recent study,14 we asked whether harnessed bees can learn the sign of reward variations so as to subsequently adjust their PERs. We used an experimental design similar to that of our initial experiment with free-flying bees.12 We first trained bees by coupling the stimulation of one antenna with either increasing, decreasing or constant reward levels offered to their proboscis throughout consecutive learning trials (Fig. 3A). We then recorded the bees’ PE reaction-time to sucrose stimulation of the antenna in the absence of reward. We found that the bees that had experienced increasing reward levels...
In our experiments, we aimed to incorporate within-animal controls into our laboratory procedure. To this end, we asked whether bees can learn side-specifically that reward level increases or decreases over time. Side-specific learning is well documented in honeybees. We developed a side-specific training in which bees were trained by coupling stimulation of one antenna with increasing reward levels and stimulation of the other antenna with decreasing reward levels throughout consecutive learning trials (Fig. 3C). Next, at different times following training, we recorded the bees’ PE reaction-time to sucrose stimulation of each antenna in the absence of reward. We found that the bees extended their proboscises earlier after stimulation of the antenna that had been linked to increasing reward levels than after stimulation of the antenna that had been linked to decreasing reward levels (Fig. 3D). Therefore, bees can also learn side-specifically subsequently extended their proboscises earlier than the bees that had experienced decreasing or constant reward levels (Fig. 3B). These results could not be accounted for by the bees’ most recent experience or the total amount of reward that they received during training. The bees that had experienced small, medium or large constant rewards showed similar reaction-times, although their last reward experience and the total amount of received reward were different (Fig. 3B). Further studies addressing neural correlates of reward memories in harnessed bees require within-animal controls. This is because recordings of neural activity are variable and, therefore, a reference from the same experimental subject is necessary for analysis of responses to any given stimulus. In a new series of experiments, we aimed to incorporate within-animal controls into our laboratory procedure. To this end, we asked whether bees can learn side-specifically that reward level increases or decreases over time. Side-specific learning is well documented in honeybees. We developed a side-specific training in which bees were trained by coupling stimulation of one antenna with increasing reward levels and stimulation of the other antenna with decreasing reward levels throughout consecutive learning trials (Fig. 3C). Next, at different times following training, we recorded the bees’ PE reaction-time to sucrose stimulation of each antenna in the absence of reward. We found that the bees extended their proboscises earlier after stimulation of the antenna that had been linked to increasing reward levels than after stimulation of the antenna that had been linked to decreasing reward levels (Fig. 3D). Therefore, bees can also learn side-specifically...
that reward increases or decreases over time. They develop both short- and long-term side-specific reward memories, and the long-term memories are extinguished by repetitive stimulation of their antennae (Fig. 3D). Also, we found that these side-specific adjustments of PE response involve an interplay between gustatory and mechanosensory input, and correlate well with the activity of muscles responsible for controlling the movements of the proboscis.15 Taken together, these findings constitute a basis on which further analyses of reward memories can be built. Such analyses will include within-animal controls and a physiological correlates of a robust behavioral measure.

The events involved in this side-specific learning are schematically shown in Figure 4. Bees learn to associate gustatory and mechanical stimulation of each antenna with either increasing or decreasing rewards offered to their proboscises through consecutive learning trials. They do this using a built-in change detector that computes differences in reward level (linked to each antenna) across feeding events. This computation leads to the formation of an internal estimate of an expected reward associated with each input side, and then, to the formation of side-specific reward memories. After training, gustatory and mechanosensory input can activate both short- and long-term side-specific reward memories. Activation of such memories leads to side differences in a honeybee’s PE reaction-time, also evinced by the activity of the muscles M17s.
relates to adjustment of a honeybee’s PER occurring after activation of reward memories. Pharmacological approaches would also prove fruitful in this context. For example, it would be interesting to evaluate the role of the octopamine (OA, a bioamine involved in associative learning, memory retrieval, and food arousal in honeybees)64–69 during formation and retrieval of reward memories.

Conclusions

The results of the above studies show that honeybees learn the sign and magnitude of reward variations and develop long-term reward expectations allowing adjustment of time/energy investment during foraging. The results also show that honeybees adjust their PE reaction-time in relation to the sign of reward variations; this form of learning involves the joint action of gustatory and mechanosensory input of the antennae, and can be side-specific. These studies constitute a basis on which three lines of investigation can be built. The first line concerns the role of reward expectations in honeybee foraging. In particular, further experiments are needed to address how the colony as a whole benefits from a honeybee’s ability to develop expectations of reward. The second line of investigation concerns the question of how reward expectations can be incorporated into theoretical accounts of individual and collective foraging. The third line concerns identifying neural substrates involved in the development of reward memories in honeybees. Progress in these three lines of investigation will bring together behavioral, theoretical and physiological data to better understand the role and underlying mechanisms of reward memories in honeybees.

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