HEATHER BELL

Honeybees use the 'waggle dance' to convey information about the location of a food source to a hive.

ANIMAL BEHAVIOUR

Nested instincts

The many levels of bee behaviour offer insights on everything from population dynamics to molecular changes.

BY LAUREN GRAVITZ

Western honeybees (*Apis mellifera*) live in highly complex societies, running nurseries and coordinating food searches that can take them kilometres away from their hives. And they do it all without leadership. "It's like the lights are on but nobody's home," says Gene Robinson, an entomologist studying bee genomics at the University of Illinois Urbana-Champaign. Despite the presence of a queen bee, the hive is not quite so autocratic as her title suggests: a queen's primary function is to lay eggs, which develop into the male drones and female worker bees that populate the colony.

Honeybees' leaderless organization provides researchers with insight into unrelated systems that have similarly decentralized control — such as the brain or the stock market. "They are compelling models for the study of social life and social behaviour," Robinson says. "They live in highly complex societies that show extreme forms of integration, cooperation and communication."

A huge part of the honeybee's appeal is that researchers can study the insect's behaviour at every level of biology, from their overall social structure to the minutia of genetics and epigenetics — as well as the interplay between these levels. Only with this complete picture can researchers hope to understand how such simple insects coordinate their behaviour so precisely and with such complexity. "The bee is well suited to address these questions at different levels of biological organization," Robinson says. Every aspect of the hive can be studied to better understand different biological systems, from how genes and epigenetics affect a single bee, to how individual behaviour can affect dynamics of the entire hive. "We have a nested Russian-doll model," says Robinson. Each doll can be studied as a separate entity yet fits neatly into the whole. And with this model, researchers hope to tease out the many layers of a complex system.

HIVE MIND

The outermost doll — the one that faces the world — is the hive itself. Bees manage their colonies by means of specific divisions of labour in which worker bees specialize in different roles. After emerging from their pupae, worker bees typically spend their first few weeks inside the hive, caring for the larvae and performing other housekeeping tasks. After that, they graduate to become foragers, and spend the next few weeks searching for pollen and nectar then sharing the locations of these treasures with the rest of the hive. The transition from hive bee to forager is not dictated by age alone — a hive has to maintain balance among its types of workers, and so bees can speed up, slow down, or even reverse this process as necessary. "If you have old forager bees in a colony, their presence inhibits young bees from becoming foragers," says Andrew Barron, who studies the neurobiology of bee behaviour at Macquarie University in Sydney, Australia. "Without them, younger bees transition faster."

Immature bees are less-effective foragers than are older bees. Barron's work has shown[1] that bees that began foraging before they were two weeks old spent less time outside the hive and went on fewer foraging flights, yet spent more time on each flight. And the younger the bee, the less likely it was to survive beyond 30 minutes outside the hive.

Thus, the hive maintains a balanced ratio of forager bees and hive bees. But if stressors (such as disease or pesticides) kill foragers at too high a rate, younger and younger bees enter the foraging force. When too many bees begin to forage prematurely, the amount of food brought back to the hive declines. And, as fewer foragers survive, new workers mature even earlier, creating a vicious cycle that can cause the colony to collapse. "The colony has a tipping point," Barron says. Quickly replacing foragers with young bees allows a hive to buffer stressors up to a point, he explains. "When that buffer is exhausted, the colony is in dramatic trouble."

The actions of individual bees can substantially alter the health of the hive. Because a hive usually has several available food sources, a colony has to allocate foragers appropriately. One of the honeybee's most distinctive behaviours is the waggle dance: a series of movements, performed by a forager on her return to the hive, that convey information to all nearby bees about the direction, distance and quality of a food source.

LANGUAGE OF THE DANCE

Because each food source varies in quality over time, bees also need a way to communicate when it is time to stop visiting a site. To convey this message, bees have developed another dance move that serves as a stop signal: a bee butts her head up against a waggle dancer and vibrates at just the right frequency to halt the waggler in her tracks. James Nieh, who studies bee behaviour at the University of California, San Diego, published his findings[2] on the stop signal in 1993. He has since determined that the bees giving these stop signals are foragers who have already been to the place the waggle dancers are promoting and discovered it to be less than ideal: perhaps the food is gone, the site is overcrowded or the bees were attacked.

In a poster at the Animal Behavior Society meeting in 2011 in Bloomington, Indiana, Nieh described how attacks by spiders, wasps and even a lab-made 'robo-predator' (forceps attached to a spring and activated by a switch), all elicit stop signals from forager bees. The same motion can also help the colony in choosing a new nest site — the fewer stop signals given to dancers describing a site, the more appealing the location. "It's a complex signal used in complex ways in very different circumstances," Nieh says. "Yet the effect of the signal in all these instances is the same: it causes all these waggle dances to stop."

Echoing Robinson's nested-Russian-doll comparison, Nieh describes the signals between individual bees as being analogous to the communication between brain cells. Just as neurons can excite other neurons, the waggle dance acts as an excitatory signal to stimulate action and foraging. And the stop signal acts much like one neuron inhibiting the signal of another. Which in effect, he says, "is how the entire brain or the entire colony achieves a complex decision".

MARKING LINES

It is in the DNA where the most fascinating — and the tiniest — of the Russian dolls sits. This is a realm that researchers are probing even deeper. Benjamin Oldroyd, who studies the behavioural genetics and evolution of honeybees at the University of Sydney, describes how bees engage in a battle of the sexes. Each queen bee mates with upwards of 20 drones, usually within a period of just a few days, and then she stores the sperm for use throughout her lifetime.

Anything that gives one male's daughters a greater chance of becoming a queen or a reproductive worker enhances his genetic legacy. But to ensure reproductive harmony among her worker daughters, a queen should ideally be able to thwart any such attempts at manipulation by the drones. Given these opposing strategies, males and females within any given bee subspecies typically evolve together to reach an equilibrium in which neither sex has a distinct genetic advantage.

Cross-breeding two bee subspecies can help to show the extent of the evolutionary changes in sexual one-upmanship. In a study[3] published in 2013, Oldroyd crossed two subspecies of African honeybee: the Cape honeybee (*Apis mellifera capensis*) and the African (or killer) honeybee (*Apis mellifera scutellata*). The female *A. m. capensis* typically has more ovarioles (components of an insect's ovaries) than do female *A. m. scutellata* bees. In theory, when the two subspecies are crossed reciprocally, it should not matter which subspecies is the father: offspring from both crosses should have the same number of ovarioles and hence the same reproductive capacity. But that was not what he found. Rather, one group of daughters had a distinct advantage: those with an *A. m. capensis* father had about one-third more ovarioles than did those with an *A. m. scutellata* father, indicating that the *A. m. capensis* males were employing a trick that extends beyond pure genetics to improve their daughters' fertility.

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Oldroyd thinks he knows how the *A. m. capensis* bees are gaining the advantage. "It suggests that males are putting epigenetic marks in their sperm to try and increase the genetic success of their offspring," he says. Epigenetic alterations are chemical tags such as methyl groups that are added to or removed from genes to turn them off or on. The honeybee was the first insect found to have a fully vertebrate-like methylation system, a fact that has led many researchers to hunt for ways in which bees may be using epigenetics to help further the success of their own lineage. If, as Oldroyd suspects, *A. m. capensis* males are epigenetically tagging the genes in their sperm in a way that enhances their daughters' fertility, then these are tags that the *A. m. capensis* females are able to counteract, but the *A. m. scutellata* females cannot.

Oldroyd is looking for additional evidence that this process involves an epigenetic mechanism. In a more recent experiment, not yet published, he removed an *A. m. capensis* queen from her hive, which led female *A. m. capensis* worker bees to start laying eggs. He found that these single-parent worker eggs have more methylation in their genome than *A. m. capensis* dual-parent eggs. This indicates that there are epigenetic mechanisms in play. Oldroyd says that he still needs to find the specific genes that are being methylated and tease out the effects, but is hopeful that he will be able find proof of the first epigenetic battle of the sexes in insects.

Examining layer after layer of the honeybee Russian doll gives researchers more than just insight into bee behaviour. Within the hive, says Robinson, are "all the traits that are important to us in understanding complex systems, whether our own society or our own bodies". ■

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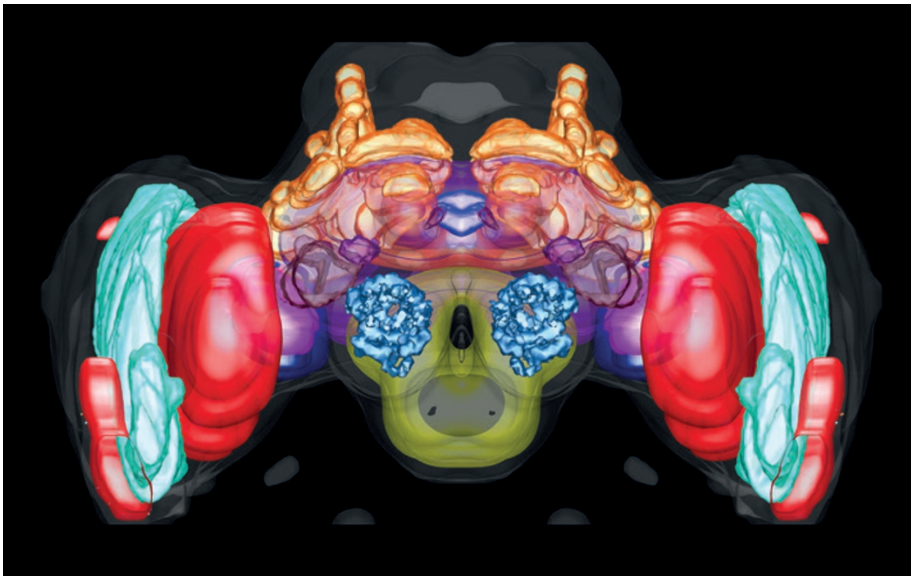

ANGELIQUE PAULK

Just like the hive, the honeybee brain has decentralized control.