What Is A Pheromone?

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Eighth In A Series . . . Examine How These Materials Are Used In The Hive

Classroom

Honey bees use a number of method to communicate. In this lesson we will start to explore the role of pheromones in honey bee communication and biology. The term pheromone is based on the Greek word *pherein* (to transport) and *hormone* (to stimulate). It refers to chemical messengers that move outside the body of an animal and result in a change in another animal of the same species. Those changes may be hormonal or behavioral. Pheromones cause animals to aggregate (group together, such as when feeding), sound an alarm (when under attack), mark a territory, release a behavior, mark a trail (as in ants), finding mates (sex), establish dominance (by royal individuals), recognize closely related individuals (to prevent mating and inbreeding), and calming (in mammals).

All major animal groups have developed pheromones, although the data on birds is limited. A common example in humans is the coordination of menstrual cycles among women, especially those living in close proximity, and who respond to the pheromone in sweat. The perfume industry is no stranger to the power of chemical signals in sexual attraction.

Dr. Mark Winston's book *The Biology of the Honey Bee*, (Harvard University Press 1988) contains a concise summary of the basic bee pheromone biology. Elsewhere Winston and his colleagues have written that there is *a remarkable and unexpected complexity in social insect pheromone communication, particularly for honeybees.* Dr. Dewey Caron's textbook *Honey Bee Biology and Beekeeping* (Wicwas Press 1999) dedicates a full chapter to chemical communications in the honey-bee colony.

After the beekeeper worked the hive, these worker bees are scenting the inner cover with their Nansonov glands exposed and fanning their wings to spread the odor of the pheromone to to bees searching for the entrance of the hive. Note how the bees have extended their legs, lowered their heads and raised the tips of their abdomens. Several clearly show the exposed Nasonov glands at that tip.

John Free *(Pheromones of the social bees*, Comstock 1987) states that honey bees have 15 glands that produce a wide range of chemical compounds, all working as chemical messengers between the queen, drone, worker and laying worker and other bees in the colony. Because these chemicals occur in different mixtures, considerable diversity of behavior is possible.

Pheromones produced by worker bees

Workers produce the **Nasonov pheromone** to assist with communication. They are a group of air-borne chemicals that include:

Geraniol
Nerolic acid
Geranic acid
(*E*)-citral
(*Z*)-citral
(*E-E*)-farnesol
Nerol

These chemicals are produced in the Nasonov gland positioned at the dorsal tip of a worker bee's abdomen. Newly emerged bees produce little of the chemicals, but their production level increases with time and reaches a peak at four weeks, or about the time of foraging activity. When the worker exposes her Nasonov gland, she fans her wings to spread the molecules in the air. Using these chemicals, bees perform these activities –

1. At the hive entrance bees release the pheromones to **guide returning foragers** and workers making orientation flights. Beekeepers often see this behavior during and after a session with the bees when the colony has been disturbed, combs moved, and the bees otherwise disrupted. Bees isolated from the rest of the hive may be seen scenting.
2. When bees are working on forage that lacks a strong natural order, as seen in a feeding station where only sugar syrup is provided, bees use Nasonov pheromone to **mark** the forage that lacks a strong odor. They also

use where the bees are collecting water. This assists incoming forgers find something that does not have a natural odor. It is very unusual to find bees scenting on natural flowers.

3. During swarming, these Nasonov chemicals are critical in the task of **keeping the swarm organized**. During their departure from the hive, while on-route to the temporary regrouping area, and then while on the way to the final nest site, bees in a swarm use the chemicals to keep the group together. They also use it to mark the location of the temporary resting site as well as the entrance of the cavity that the have selected to be their new home. It is thought that the queen herself uses these molecules to orient within the swarm, and her royal pheromones undoubtedly work in conjunction with the Nasonov compounds to keep the swarm intact. Beekeepers know that a swarm without a queen will eventually return to its initial nest.

Another worker-generated compound is the **footprint pheromones**. These odors are left by worker bees at the entrance of the hive and on flowers. It is unclear whether these compounds are produced by the Arnhart glands in the tarsal segment or are produced elsewhere on the bee's body and deposited by the feet. When the footprint pheromones are found at feeding sites, more bees are attracted to the location. They are believed to combine with the Nasonov substance to produce a chemically attractive hive entrance for returning forager and bees on orientation flights. This is a synergistic combination of two different compounds for an improved outcome.

Beekeepers are familiar with the next group of worker-bee produced pheromones, the sting-produced compound (Z)-11-eicosen-1-ol. This is both an alarm compound and attracts other worker bees. Another odor, thought to be produced by the dorsal (top) surface of the abdomen, is called the forager-marking pheromone.

Workers produce a variety of odors employed to initiate the **alarm and coordinate colony defense**. One compound is 2-heptanone, produced in the mandibular glands of workers, while the remaining compounds are produced within the sting structure, most likely in the membranes at the base of the sting lancets. These molecules work together to tell other bees that the colony is under attack and to mark the location on the attacker where the warning sting was inserted. The stinging experience is complicated, and combines chemical signals with movement behavior to result in a full response. 2-heptanone does not illicit a strong response. Another compound, isoamyl acetate (also labeled isopentyl acetate or IPA) is 20 to 70 more times likely to result in a sting behavior response. Add to these the chemicals butyl acetate, 1-hexanol, n-butanol, 1-octanol, hexyl actate, oxtyl acetate, n-pentyl acetate and 2-nonanol. These are highly volatile low molecular weight chemicals. They are released into the air and attract other bees, often resulting in defensive and stinging behavior.

Isoamyl acetate and other defense chemicals are released when a bee stings and the sting membranes are torn. Then it attracts other worker bees to the defense site and increases the intensity of the defense response. A strong banana-like odor should instruct the beekeeper to close the hive and let the bees settle for a day or more, or be faced with multiple stinging attempts. The use of smoke interferes or masks the bees' alarm pheromone.

2-heptanone is used to deter potential enemies and robber bees. It may be used as an anesthetic to paralyze intruders, allowing the bees to remove paralyzed individuals from the hive.

Forager pheromone

Older forager bees release **ethyl oleate**, often detected by nurse bees by antennal contact on the body of the forager. This compound slows the development of nurse bees. This is an example of a **primer pheromone**, and it keeps the age ratio of nurse bees to forager bees in a efficient balance.

Pheromones produced by brood

Developing bees and pupae produce a **brood recognition pheromone** that inhibits worker bee ovary development, and helps nurse bees discriminate between drone and worker brood. It draws nurse bees to the brood, and is used by beekeepers, often unknowingly, as a means of making new colonies.

The compound at work here is a 10-compound of fatty-acid esters which help regulate the adult caste rations. For a long time it was thought that queen pheromones inhibited the development of ovaries in worker bees, but now, instead, evidence suggests that the brood pheromones are responsible for this inhibition so workers do not lay eggs. It correlates with the observation that laying workers appear in a colony about month after a queen is removed and a replacement fails to replace her. This means the brood from the original queen has all emerged and is none left to suppress ovary development in workers.

In 1996 an artificial brood pheromone was developed by a team of researchers lead by French researcher Yves Le Conte.

Note: We will continue the discussion of other hive pheromones in the next article, including the pheromones produced by the queen bee.

Class Activities

Observation hive – Make arrangements to borrow an observation hive or set one up with your bees, perhaps from a nucleus. Make sure you include the laying queen bee, preferably marked for easy identification. In the classroom, have students observe the queen for shifts of two minutes, recording what the queen is doing during these time periods – inspecting cells and laying eggs, walking, resting, etc. As she does this have students count the number of worker bees that appear to be attendant bees, doing this four times, at 30 second intervals. Attendant bees are those that are touching the queen, feeding her, and otherwise somewhat focused on the work of the queen and nothing else. Repeat these observations on at least two different days. Have each student prepare a report of their observations, keeping in mind that each may have observed a different period of the queen's daily activity cycle.

Field session – Produce a temporary package of screened cage of bees from an existing hive or nucelus. Find the queen and cage her, without workers, in a plastic or wood and screen cage. Put her in the cage with the worker bees, but secure in the cage so she cannot get out. The next day, remove the queen cage and fasten it to an artificial tree, or cross of wood, in the open air. Shake the worker bees at on the ground at the bottom of the tree and observe their searching and scenting behavior they use to find the queen. Put some workers on the queen cage to observe the scenting at a closeup basis. After a period of time the bees will have created an artificial swarm around the queen, and students should study the movement of bees in the swarm to see how it is structured by the bees.

Set up a second wooden tree and carefully remove the queen in her cage and gently brush off ALL the attendant bees on the cage. Move the queen to the second tree located 15 to 20 feet away from the first tree, and expect to spend several minutes watching the bees break the cluster, search for the queen, find her, start scenting her new location, and then regrouping to the second tree.

As a group examine the changes to the bees and how they deal with the loss of their queen. A review of the pheromones involved in this behavior is part of the session.

If possible video record this exercise, and try to get as many closeup views as possible. Have the students compile the video into a report, to share with other students and family. BC

Vocabulary

pheromone, alarm, volatility, aggregation, releaser pheromone, Nasonov gland, footprint pheromones, brood pheromones, 2-heptanone, isopental acetate (isoamyl acetate), swarm organization, breaking behavior, queen retinue, egg laying behavior, royal dominance, trail pheromones, repellency, primer pheromone, ethyl oleate, nurse bee to forager bee ratio

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