Understanding Queen Pheromone?

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

Last month we reviewed the pheromones of the honey bee colony. In the field section, we described an activity to study the workers' attraction to the queen pheromones. Now it is time to concentrate on the pheromones of the queen honey bee.

Classroom

What are the queen's pheromones?

Information about the queen bee's pheromones is growing, and we will concentrate on these chemicals and their impact on bees. The queen mandibular pheromone (QMP) is often called the *queen substance*, a term given to it by Dr. Colin Butler in 1954. The primary component is 9-oxydec-2-enoic acid, commonly called 9-ODA. This chemical was isolated from the mandibular glands of queen bees. These glands are connected by a duct that connects to the mandibles of the queen. There are two glands, one for each mandible.

The chemical we call queen substance is actually 17 different compounds that have been isolated from mandibular glands. Dr. Dewey Caron, in his textbook *Honey Bee Biology and Beekeeping*, lists five molecules as the "most important". These five molecules work together, and the removal of any one of them cuts the level of "excitement" by half. The first three molecules are acids, and the last two are aromatic compounds:

9-ODA	9-oxydec-2-enoic acid
9-HDA	cis-9-hydroxydec-2-enoic acid
9-HDA	trans-9-hydroxydec-2-enoic acid
HOB	methyl-p-hydroxyenzoate
HVA	4-hydroxy-3-methoxyphenylethanol

Queen without a well formed retinue of worker bees, common when a queen is active on the comb.

These molecules and their role as pheromones were discovered in a series of research projects conducted since 1961. Work continues to discover other ways these molecules are utilized as part of the queen pheromone blend.

Queen retinue pheromone

A laying queen honey bee apparently performs two major functions in the colony. Ideally, she will oviposit about 200,000 eggs into worker and drone cells over a year, serving as the biological mother of all the bees in the hive, passing her genes to her daughter queens and her drone sons. Second, she is the producer of pheromones from glands in her body.

In 2003 we learned more about the concept of the queen retinue pheromone, which allows other molecules from the queen's body to contribute to the behavior we see in bees that visit, groom her, feed her, and receive her pheromone in the process. Most beekeepers have watched the queen when surrounded by her retinue of nurse worker bees. This occurs when the queen is resting from egg laying. During active egg laying the retinue does not exist or is poorly defined. The bees in the retinue are constantly changing, some only make brief contact with the queen with their antennae, during which pheromones are passed from queen to worker. These workers are then in an "excited" mode, carrying the pheromone throughout the hive to other workers. This spreads the chemicals throughout the colony so all bees are aware of the queen's presence even though most of them have not had physical contact with her.

Other retinue workers lick the queen's body and obtain pheromone, while others feed and others remove her feces. The queen rectum produces pheromone and pheromone-loaded feces are found in other animals. This includes the only mammal with a queen, one that suppresses her daughters reproduction by pheromones in her feces, the naked mole rat of Africa.

At Simon Fraser University, researchers measured the amount of these different chemicals produced by mated, laying queens. They call this blend of pheromone a *queen equivalent* (Qeq):

Chemical	**Amount**
9-ODA	~200 micrograms
9-HDA (both forms)	80 micrograms
HOB	20 micrograms
HVB	2 micrograms

A queen secretes one Qeq every 24 hours. She moves the pheromone to other parts of her body by her self-grooming and by worker grooming. Virgin queens produce significantly less 9-HDA, HOB and undetectable amounts of HVA. As a result, virgin queens are much less attractive to worker bees. Once mated and laying eggs, the queens are fully attractive to the bees in the colony. This helps us understand how virgin queens are relatively free to move around the colony until they are mated. Since they do produce *some* of the Qeq, beekeepers must treat them much the same as a mated queen; they cannot be dropped into a foreign colony and be expected to survive.

Genes control queen pheromone production

Queens differ in their queen mandibular pheromone due to different genes they possess. Breeders selected for high or low producers of the queen mandibular pheromone and the high QMP strain produced nine times more pheromone than the low strain.

In the strain with low QMP production, the bees still responded to the queen in a normal manner, suggesting there are other molecules produced by the queen that added to the queen retinue response by worker bees. Pheromones were moved from the head to the tergite gland in the top of the queen's abdomen.

Four new retinue molecules

As other researchers examined the queen retinue pheromone, they found four new molecules that work in synergy with QMP to excite workers. The nine compounds do not combine to provide the same retinue response of queen extract at higher doses, suggesting that still more molecules are yet to be discovered. The four new molecules are:

methyl oleate — methyl (A)-octadec-9-enoate

Coniferyl alcohol — (E)-hydroxy-3methoeyt-phenyl)proop-2-en-1-ol

Heyadecane-1-ol

Linolenic acid — (Z9,Z12, z15)-octa-dexa-9, 12, 15-trienoic acid

These pheromones are produced in different glands in the queen's body, and their production may be influenced by the metabolism of each gland. Some molecules are found on the queen that previously were found on honey bee brood, suggesting that the queen works to control worker physiology and behavior,

Notice the worker bees antennal closeness as they detect and receive pheromone from the queen bee.

and regulates retinue bees in the amount they lick, antennate, and groom her. This implies that the queen controls more of the behavior of the bees than previously thought, at least in getting someone to pay attention to her. *From Bee Sex Essentials*

Field Activity

Set up two nucleus hives in four or five frame colonies, if you have them. At least one frame should be worker brood. The second frame should be a frame of food, pollen and honey. Fill the remainder of the hive with empty drawn comb or frames with foundation or starter strips.

In one of the nucleus hives, install a laying queen in a cage with a candy release. Leave the second hive queenless.

On the fourth or a later day, examine each hive using no smoke or just a minimal amount to settle the bees. Examine and document the behavior of the bees for each of the following:

Number of bees that fly into the air (estimate)

The sound of the colony: Quiet to Noisy

Behavior of the bees on the comb: Quiet on the comb or running on the comb.

Do the bees drip off the comb or fly into the air at the same rate as you inspect each colony?

Have each student write a paragraph describing the differences between the two colonies, if any. Encourage their use of expressive language to best and most accurately describe what they have observed. Share the essays

with the rest of the class.

Observation Hive Activities

Establish two observation hives. In one, install a newly emerged virgin queen. In the second observation hive, install a mated queen that has been laying viable worker eggs for at least one month. This could be a queen from spare nucleus colony you have established during the season.

In a series of timed observations, compare the two queens for their general behavior patterns. Once the bees in the observation hive have settled, record the comb area that the two queens cover in a two minute period. This may be expressed by the travel distance the respective queen covers in the two minute period, or the percentage of comb that the queen covers during this time period.

Also observe the behavior of other bees toward the queen, by counting the number of worker bees that make contact with the queen with their antennae. We think that this antennal contact is critical to the spread of certain pheromones by the mated queen, so it is useful to see how many workers respond to the virgin queen.

Instruct the students to track one or more of the bees that antennate the queen as they leave the retinue and move about the comb. Are they different from other bees? Do they share food contents? Do other worker bees antennate them? Look for indications that these bees are messenger bees, spreading the queen's pheromones as they work the hive in an excited state.

Have each student describe the behavior of each queen, using as many descriptive words as they can. Then make comparisons between the two queens. For example, one might find that the virgin queen is 'fast' on the comb, while the mated queen is 'slow' or 'slower.' Have fun with these comparisons.

Finally, have each student study the laying queen as she searches for empty cells to deposit eggs. Have them count the number of attendant or retinue worker bees around the queen every fifteen seconds, and note the behavior of the queen during this process. Is the queen laying eggs, walking on the combs, quiet and resting or what? Again, let the students develop the descriptive language for these observations.

At the end of the session, list the words and phrases the students have developed to describe the behavior they have seen. Discuss the words that are solid descriptive terms and those that may project some value or anthropomorphic view of the queen bee. Turn this lesson into a unit on making good descriptive statements rather than opinions or value judgements. BC

Vocabulary

queen retinue pheromone, egg laying behavior, queen mandibular pheromone, QMP, queen substance, 9-ODA, two functions of the queen, excited mode of worker bees, antennation, queen's presence queen rectum pheromone, naked role rat, queen equivalent, genetic control of queen pheromone production, nucleus colony setup, sound of a hive, bees dripping off combs, behavior of virgin queens, behavior of laying queens, scientific observations, descriptive language, anthropomorphism

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