

Honey Bee Brood Pheromone for Enhanced Pollination Activity

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Objectives:

To develop honey bee brood pheromone for enhanced pollination activity that is 1) chemically stable, 2) uses less expensive technical grade chemical compounds, and is dispensed through 3) a long-term delivery device.

Interpretive Summary:

In 1989 the discovery of a 10-component blend of chemicals communicating the presence of honey bee larvae called brood pheromone was reported by Le Conte et al.⁽¹⁾ Nine years later Pankiw et al.⁽²⁾ discovered that brood pheromone stimulated increased honey bee pollen foraging behavior. At that time brood pheromone's potential as a practical tool for almond growers and beekeepers was recognized; however, it would take an additional 9 years for this potential to be realized while more basic research summarized in Table 1.

Table 1. Brood pheromone is multifunctional releasing many forager behavioral responses and priming physiological responses in pre-foraging worker bees.

Releaser Responses	Primer Responses
Increases number of pollen foragers up to 150% ⁽²⁻⁹⁾ .	Recruits younger bees into foraging. ^(4, 6, 8, 10)
Increases pollen load weight returned ⁽³⁾ .	Primes hypopharyngeal gland development and protein production in cage environments ^(6, 8, 11, 12) and in colonies in the summer and winter ^(6, 12-14) .
Increases number of pollen forager trips per unit time ⁽¹⁵⁾ .	Increases colony growth rate in the summer and winter ^(6, 12, 13) .
Increases number of pollen grain extractable from the bodies of non-pollen foragers ⁽³⁾ .	Increases winter colony protein supplement consumption ¹⁸ .
Increases number of pollen & non-pollen foragers ⁽³⁾ .	Partially inhibits worker ovary development in queenless environments ⁽¹⁶⁻¹⁸⁾ .

The principal problems hindering brood pheromone development were chemical instability resulting in a rapid loss of biological activity, the high cost of highly purified reagent grade chemicals used to formulate the pheromone, and a long-term delivery device to replace impractical daily applications. In a collaboration beginning in 2004 we proceeded to solve each problem resulting in development of a technology called SuperBoost. SuperBoost is a slow-delivery device containing brood pheromone that is chemically stable and remaining biologically active after long-term storage at hive temperature. SuperBoost is made with technical grade compounds that reduced the cost of formation by at least 93% compared to formulation with highly purified reagent grade compounds. The technical and reagent grade formulations of brood pheromone are not significantly different in their effect on stimulating increased pollen foraging. SuperBoost stimulated increased pollen foraging activity and colony growth for a 6 week period that was consistent with previous studies summarized below and in Table 1.

Honey bee workers show behavioral development where in general, younger pre-forager bees perform tasks found in the nest and older bees forage for food outside the nest. In a honey bee colony, age related behavioral development results in a division of labor. For example, brood rearing in honey bees is accomplished by the combined labor of nurse and forager bees who directly and indirectly provision larvae, respectively. Pollen foragers collect pollen from available plant sources then return to the nest and deposit their loads of pollen directly into cells. Stored pollen is consumed by nurse bees that use the proteins derived from the pollen to produce proteinaceous hypopharyngeal gland secretions that are fed to developing larvae. It is through nurse bees that larvae are the principal consumers of pollen in a colony. Increasing the amount of larvae in colonies, or brood pheromone, increases the number of pollen foragers and pollen load weights returned ^(2, 5, 19-21). Brood pheromone communicates amount of larvae present in a colony. Larva number is estimated by amount of brood pheromone and adult worker behaviors and physiologies adjust accordingly.

Brood pheromone affects foraging behavioral development⁽¹⁰⁾, such that some workers of the same age in the same colony forage at significantly younger ages and some may

be delayed significantly longer as nurses compared to workers of the same genotypic background and age cohort reared in control colonies⁽⁴⁾. Treating colonies with brood pheromone daily for 4 to 5 weeks results in sustained significantly increased pollen foraging and increased rate of colony growth^(6, 13). Queens in the brood pheromone treatment laid more eggs, were fed longer and were less idle compared to controls⁽¹⁴⁾. Significantly more time was spent in cell cleaning by the bees in pheromone treated colonies⁽¹⁴⁾. Brood pheromone regulated queen egg-laying rate by modulating worker-queen interactions and nurse bee rearing behaviors.

Larvae and their chemical cues called brood pheromone appear to be principal organizers of colony activity. Therefore it is not surprising that brood pheromone is known to regulate more honey bee worker behaviors and physiological systems than any other honey bee pheromone⁽⁴⁾. It should not be surprising that so many colony activities are centered on its young. Colony growth and reproduction are dependant on the queen as an egg layer and amount of young in a colony. For growth and reproduction to occur, workers must appropriately respond to the number of larvae in a colony. Brood pheromone has been demonstrated to have profound effects on honey bee individuals and colonies. Colony organization is centered on brood rearing such that larvae and their chemicals appear to regulate virtually every aspect of colony life.

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