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Neuroethological study of pheromonal sex communication in honeybee drones

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During the mating season, male honeybees, the drones, gather in drone congregation areas (DCA). When a virgin queen enters the DCA, drones are strongly attracted by olfactory cues (sex pheromones). The mechanisms allowing drones and virgin queens to find the DCAs are still unclear, and the possible existence of a drone-produced pheromone has been proposed, but not demonstrated. We thus investigated innate odor preferences of drones under controlled laboratory conditions using a locomotion compensator. First, we tested the behavioral responses of drones to 9-oxo-2-decenoic acid (9-ODA; major component of the virgin queen odor bouquet) and to queen mandibular pheromone (QMP; pheromonal mixture specific of mated queens, including proportionally less 9-ODA). Drones were attracted to 9-ODA, but not to QMP. Next, we investigated the potential attractivity of drone-derived odors. We found that honeybee drones are indeed attracted to the odor bouquet from live drones but not to the bouquet of workers. Such attraction between consexuals may play a role in the formation and maintenance of DCAs. The olfactory system of honeybee drones is highly-tuned to queen pheromones. The drones from almost all known honeybee species show a strong attraction to 9-ODA. However, the composition of the mandibular gland pheromone differs clearly between species. How did the drone olfactory system evolve to process these compounds? We performed a comparative neuroanatomical study of the drone antennal lobe (AL; primary olfactory center) to identify pheromone-specific processing units (macroglomeruli). Our results suggest a complexification of AL structure from so-called ancestral species (dwarf and giant honeybees, like *Apis dorsata*) to the most derived ones (cavity-nesting honeybees, like *A. mellifera* and *A. cerana*), with more macroglomeruli. In addition to numerical differences, we also find clear differences in macroglomerulus localization between species. Implication of these results for our understanding of bees sexual behavior will be discussed.