

Observation Hive Marking Apparatus (OHMA): An Inexpensive Method For Mass-Marking Honey Bees In Observation Hives

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Introduction

Hagler, Mueller, Teuber, van Deynze, and Martin (2011) developed an effective way for mass-marking honey bees from multiple apiary locations. The method, however, only works with Langstroth hives. Since honey bee research is often conducted with smaller observation hives, we made modifications to the method for mass-marking bees from these smaller hives. We used this modified approach to track hive-specific foraging preferences. By using multiple hive-specific colors, we were able to count the number of bees from each hive at each experimental foraging location (Figure 1). Using observation hives allowed us to easily track hive-specific micronutrient preferences (Bonoan et al., in revision).

Dust-Marking Insects

Scientists have been marking insects since the 1920s, beginning with simple paints, dyes, and stains (Hagler & Jackson, 2001). Recent developments in insect marking include protein marking (e.g., ELISA)

(Hagler & Jackson, 2001), genetically engineered marking using transposable elements (Hagler & Jackson, 2001), and radio-frequency identification (Schneider, Tautz, Grünwald, & Fuchs, 2012). Such recent developments are more accurate but are expensive and not always necessary.

Here, we describe a method using Day-Glo's fluorescent ECO Pigments (Day-Glo Color Corporation) which are visible to the naked eye (Figure 2), and free of harmful chemicals (such as formaldehydes and heavy metals). ECO Pigments come in a variety of colors – thus, multiple hives can be individually marked – and do not have any negative effects on honey bees (Hagler et al., 2011). Dust-marking can also be done by puffing the insect with a cloud of colored dust or by tumbling the insects in a dust-filled container (Hagler & Jackson, 2001); however, both require handling of the insects. In both this modified method and the Hagler et al. (2011) method, the insects self-mark, and thus are not handled by an experimenter.

The Method

Our observation hives are housed in an enclosure with an exit tube running through the wall to allow bees to forage freely (Figure 3). Before making the observation hive-marking apparatus (OHMA), the bees must have a landing strip. The exit tube needs to be flush with the outside wall so that the OHMA can be attached to the exterior wall on either side of the tube (Figure 3).

Other than the marking powder, all materials to create the OHMA are available at a hardware store. The device itself is constructed with vinyl mesh, a hot glue gun, and heavy duty Velcro (Scotch Extreme Fastener, 1" × 10'). First, we created a mesh satchel for the powder by cutting a 4 cm × 8 cm piece of mesh, folding the mesh in half, and hot gluing the two ends together. At this point, we had a 2 cm × 8 cm mesh satchel with an opening on top for loading the powder. Then, we cut two pieces of Velcro (about 1 cm × 2 cm) and hot glued one piece to each end of the mesh



Figure 1. Mass-marked honey bees from the yellow, pink, and orange hives foraging for micronutrients at our experimental foraging area. Photos: Rachael E. Bonoan.

satchel. Next, we cut two more pieces of Velcro (about 1 cm × 2 cm) and stuck them to the wall on either side of the exit tube. We used small picture-hanging nails to further secure the Velcro in place. The Velcro allows for the OHMA to be applied and removed multiple times.

When it was time to mark the bees, each satchel was filled with about 15 grams of marking powder. The satchel was then secured onto the Velcro so it blocked most of the exit hole. This requires the bees to brush up against the mesh, and thus get dusted with the colored powder, as they leave and enter the hive. The bees quickly adjust to the constrained exit/entrance tube. This is where the landing strip is important – the bees require a place to land and then crawl into the hive under the OHMA, or else the bees cannot get back in. At the end of the experimental period, the OHMAs can be removed to allow the bees to come and go more freely.

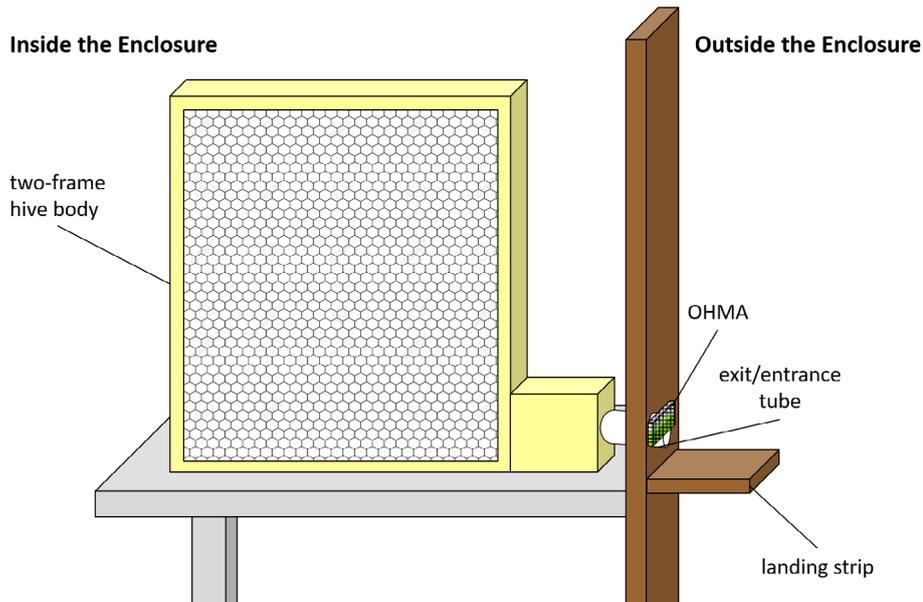
Since we were only interested in identifying bees by hive for a short foraging time, this method worked perfectly (Figure 1). The foraging area where we observed the marked bees was about 100 m away from our observation hives; bees remained marked throughout observations. Although 100 m is not far for a forager, neighbors with gardens and students on



▲ **Figure 2.** A forager dusted with yellow marking powder.
Photo: Rachael E. Bonoan.

campus observed marked foragers (anecdotal evidence) beyond 100 m. The bees tend to mostly clean themselves off before they return to the hive; however, there are parts of their body they cannot reach, and hence remain marked. In some cases, the pigment did make it into the wax comb but there was never any discoloration of the stored honey or pollen.

The OHMA can be used to mass- and self-mark insects wherever there is a small exit/entrance hole. For example, OHMAs can be situated near floral visitation sites, insect bait stations, insect traps, and at natural nest entrances (Hagler & Jackson, 2001). With honey bees in particular, OHMAs can be used to investigate nuanced questions regarding honey bee behavior both inside and outside the observation hive (Table 1, and references therein). Outside the hive, this method allows for identification and tracking of unique hives. Inside the hive, this method allows for an easier way to visually follow bees upon their return to the hive (Ikeno et al., 2008).



▲ **Figure 3.** Experimental setup modeling the observation hive and OHMA.

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Table 1. Types of questions that can be investigated using the OHMA.

Behavior	Questions OHMA would allow	Reference(s)
Swarming	If there are multiple swarms that result from one hive, do the swarms ever combine?	(Allen, 1956; Fefferman & Starks, 2006; Rangel & Seeley, 2008)
	Do swarms from different hives combine?	
Grooming	When dusted, how long does it take a forager to groom before returning to her hive?	(Božič & Valentinčič, 1995)
	Do nectar/water foragers groom more than pollen foragers (as the pollen foragers risk losing their pollen)?	
	Do hives differ in forager grooming time?	
	Does forager grooming time affect hive health in any way?	
Foraging	Do hives exhibit different foraging preferences?	(Pernal & Currie, 2001; Steffan-Dewenter & Kuhn, 2003)
	Do such preferences change with season?	
	Do such preferences change with hive health/in-hive dynamics?	
Pollination	Which flowers have been visited by a marked bee?	(Danner, Härtel, & Steffan-Dewenter, 2014; Degrandi-Hoffman & Chambers, 2006)
	Which flowers have been visited by a certain hive?	
Communication	Where does the focal individual of a waggle dance go after the dance is through?	(Farina, 1996; Schürch et al., 2013; Tanner & Visscher, 2006)
	Does waggle dance variability differ between hives? (i.e., Are some colonies better at waggle dancing than others?)	
Guarding	How do guard bees (bees marked one color) react to foreign bees (bees marked another color) during different times of the year? Is there a seasonality to aggression/defensiveness?	(Unger & Guzman-Novoa, 2012)
	How might this differ between hives?	

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