Tracheal Mite Microscopy

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Introduction

Tracheal mites (*Acarapis woodi*) are internal parasites that live in the prothoracic trachea of adult honey bees. These mites feed on the bee itself, piercing the bee's tracheal tubes. Infestation ultimately reduces the bee's lifespan and its ability to fly and can lead to population decline within the colony. Visual signs of *A. woodi* in adult bees are not unique to this parasite, and the mites themselves cannot be seen with the naked eye. Accurate diagnosis, therefore, requires dissection of adult honey bees and microscopic examination of their trachea.

Indications

Tracheal mites may not always cause obvious visual signs at the colony or individual bee level. A beekeeper may note multiple bees exhibiting k-wing (Figure 1) or see many bees crawling around near the hive entrance. Colonies with high tracheal mite infestations may show decreases in colony population, brood, and food stores. Severe infestations can lead to colony death, usually in the late winter or early spring. Dissection is warranted in a colony with trembling or disoriented bees in the late fall, winter, or early spring.

Figure 1: Honey bee exhibiting k-wing, where the wings on one or both sides are held out at an angle indicating discomfort, sometimes associated with tracheal mites and other stressors. Photo by UF/IFAS Honey Bee Research and Extension Laboratory.

SUPPLIES NEEDED

Screw-cap containers (one per colony to be sampled)

70% ethanol or isopropyl alcohol

Dissecting microscope, up to 50x magnification

Petri dish containing wax (or other pinning substrate)

Insect pins

Two pairs of micro forceps

Optional supplies (to view *A. woodi* within the trachea)

Compound microscope (100-400x magnification)

Glass microscope slides

Glass coverslips

Water

Sampling Bees

Label a sample container for each colony that you plan to test, adding sufficient 70% alcohol to cover a sample of approximately 50 bees. From each colony, try to collect about 50 older worker bees, targeting any bees that are crawling at the hive entrance or exhibiting K-wing. Older workers can also be collected from under the hive lid, from the outermost frames, or at the hive entrance.

Preparing the Sample

Sampled bees should be dissected within 90 days of collection to avoid degradation of the sample. After this time, the honey bee trachea (Figure 2) will start to darken, making it more difficult to detect tracheal mites.

Remove a bee from your sample container. Attach the bee to your pinning substrate with two insect pins, one on each side of the bee's thorax. Place the pinned bee under a dissecting microscope at 20x-30x magnification. Using micro forceps, remove the head and first pair of legs from the bee. Next, remove the first tergite of the prothorax, which is the first ring on the bee's thorax also known as the collar (Figure 3). These dissections will expose two inverted V-shaped mesothoracic tracheal tubes. See *Standard methods for tracheal mite research* (Sammataro et al., 2013) for dissection details.

Figure 2: Diagram of tracheal tubes located in the honey bee thorax, with a magnified section showing tracheal mites. Diagram by Mary Bammer, UF/IFAS Honey Bee Research and Extension Laboratory.

Figure 3: Honey bee with head and first pair of legs removed. The arrow and black line indicate the honey bee's collar. Photo by Lyle Buss (University of Florida) in Sammataro et al (2013) (http://dx.doi.org/10.3896/IBRA.1.52.4.20).

Observing the Trachea

Healthy honey bee trachea should be smooth and clear, although they may take on the white or amber color of the tissue behind them (Figure 4). Infested tracheae start to become cloudy or otherwise discolored. Moderate to heavy infestations will lead to dark patches and scarring on the tracheae (Figure 5). You may need to adjust magnification to more clearly view the exposed trachea. For light infestations, tracheal damage may not be visible; removing the tracheae and examining at higher magnification is then required for diagnosis. For instructions on this process and for alternative methods of detecting tracheal mites in honey bees, see *Standard methods for tracheal mite research* (Sammataro et al., 2013).

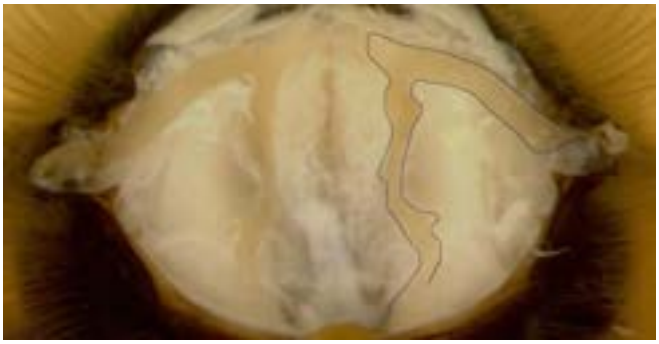

Figure 4: Figure 2: Exposed mesothoracic tracheal tubes that do not have visual signs of tracheal mites. The right tube is outlined in black. Photo by Lyle Buss (University of Florida) in Sammataro et al. (2013) (http://dx.doi.org/10.3896/IBRA.1.52.4.20).

Figure 5: One exposed mesothoracic tracheal tube showing signs of a heavy tracheal mite infestation, including cloudy tubes and dark scarring. Photo by Lyle Buss (University of Florida) in Sammataro et al. (2013) (http://dx.doi.org/10.3896/IBRA.1.52.4.20).

Interpreting the Results

Use the table below from *Standard methods for tracheal mite research* (Sammataro et al., 2013, accessed 2023) to determine if each colony is considered to have a low (<10%) or high (>10%) infestation, if more bees should be dissected, and if treatment is recommended.

No. of bees examined	Number of infested bees		
	Low infestation (stop, don't treat)	High infestation (stop, treatment)	Moderate infestation (continue sampling)
10	-	4	0,1,2,3
20	1	6	2,3,4,5
30	3	8	4,5,6,7
40	5	10	6,7,8,9
50	7	11	8,9,10

Other Tracheal Mites

Besides *A. woodi*, two other species of tracheal mites, *Acarapis externus* and *Acarapis dorsalis*, are associated with adult honey bees, although neither are known to directly harm bees. These three mites are most commonly distinguished by their location within the bee, with only *A. woodi* commonly found in the prothoracic trachea as described in this lesson.

This work is created with funding from the Veterinary Services Grant Program (VSGP) Education, Extension and Training (EET) competitive grants program of the USDA National Institute of Food and Agriculture (NIFA) (2202-04170) and the USDA NIFA Sustainable Agriculture Research and Education (SARE) Research and Education Grant program (LNC22-468).