Interesting Things I've

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Introduction

There are many maladies that can affect the health of a beehive. Their effects range from nuisance to serious problem affecting hive survival (Arbia & Babbay, 2010). Fungal and bacterial diseases include American foulbrood (AFB), European foul brood (EFB) and chalk brood (Bailey, 1983; Ashiralieva & Genersch, 2006; Heath, 1982). These diseases do not affect the adult honey bees, but profoundly affect the developing brood (Oldroyd, 2007). Protozoan pests include two *Nosema* species, *Nosema apis* and *Nosema cerena,* which infest the gut of adult bees and can cause dysentery and death (Zander, 1909; Fries, 2010). Additionally, most honey bees carry viruses that under normal conditions do not cause harm to the honey bee (Ribière et al., 2000; Nielsen et al., 2008). Gajda et al. (2021) found at least 36 different bee viruses inside of tested hives. They posit that only seven of these are likely to cause harm to the bees. Those viruses likely to cause harm are deformed wing virus (DWV), acute bee paralysis virus (ABPV), black queen cell virus (BQCV), bee virus Y (BVY), *Apis mellifera* filamentous virus (AmFV), chronic bee paralysis virus (CBPV) and sacbrood virus (SBV). Two of the most common insect pests of beehives include the *Varroa destructo*r mite and the lesser wax moth *Achroia grisella* (Oldroyd, 1999; Burges & Bailey, 1968). This case report will detail and unusual instance of *A. grisella* in a healthy beehive.

Impact of Wax Moths on Beehives

The lesser wax moth, *A. grisella*, typically inhabits and consumes abandoned or empty bee hives of honey bees (Clarke, 1986). Additionally, *A. grisella* can inhabit the stored beekeeping equipment that contains beeswax. Although *A. grisella* is referred to as the wax moth, the larvae don't digest the comb wax (Clarke, 1986). Instead, they rely on the impurities within the combs, such as bee brood, honey and pollen, for their nutrition (Egelie et al., 2015). Most commonly, they tend to feed on wax combs where brood has been raised rather than clean honeycomb or honeycomb containing honey (Clarke, 1986; Egelie et al., 2015). Even though the feeding behavior of *A. grisella* is beneficial to wild bee colonies because the larvae consume and recycle the old honeycomb of abandoned or dead bee colonies thus clearing the space for a new colony to build, it is estimated that *A. grisella* larvae cause over five million dollars of damage annually to beekeepers in the United States (Caron, 1992).

Life Cycle

The life cycle of *A. grisella* consists of four stages: egg, larva, pupa and adult. Adult females lay up to 300 eggs that hatch into larvae in five to eight days depending upon the temperature, with warmer temperature shortening the hatching time (Egelie et al., 2015). *A. grisella* eggs are creamy white and spherical. They are typically laid in cracks or other protected areas close to the food source (Egelie et al., 2015). Eggs laid in hidden and protected areas combined with the small size of the eggs causes them to frequently go unnoticed by beekeepers.

Newly hatched larvae burrow into the comb seeking to reach the midrib of the comb (Caron, 1992). Most commonly, the larvae feed on weak or stressed colonies for the next one to five months (Egelie et al., 2015). The larval stage is the only phase during which the wax moth feeds. They generally feed on wax comb containing bee brood, pollen, or honey with preference given to brood and pollen over honey or virgin comb. In cases where both the lesser and greater wax moth (*A.grisella* and *Galleria mellonella*) are found in the same hive, the lesser wax moth can be found feeding on the hive floor because it is out-competed for space by the greater wax moth. Developing larvae tunnel through the beeswax comb in search of food leaving behind a trail of silk and feces (Egelie et al., 2015). Larvae undergo a total of seven molts with most of the growth occurring during the final two instars (Egelie et al., 2015). Larvae become fully developed in an average of six to seven weeks at a temperature range of 29°C – 32°C. Fully grown larvae are gray, about 20 mm long with a dark head capsule, three pairs of legs and several body segments (Egelie et al., 2015; Caron, 1992).

Larva enter the pupal stage by spinning a tough, silk cocoon around themselves (Egelie et al., 2015). They can sometimes be found on the bottom board among the hive debris but are most commonly found anchored to the hive body or frame (Caron, 1992). The larvae chew a cavity into the wooden hive body into which the frass and debris cocoon are affixed, further contributing to the damage caused by wax moths beyond the destruction of honeycomb (Caron, 1992; Egelie et al., 2015). It can take up to two months for the adult to emerge from the cocoon depending upon the temperature with typical emergence time being about 37 days (Egelie et al., 2015).

Adult male wax moths are slightly smaller than the females. Adult moths are most active at night and live for about one week (Caron, 1992). During this time, they will mate within the beehive. Males use ultrasonic frequencies to attract females who will lay their eggs inside the hive at night. During the day, the moths are mostly inactive and hidden in dark spaces outside of the hive (Caron, 1992; Egelie et al., 2015).

Treatment and Mitigation Strategies

Treatment and mitigation strategies for wax moths includes three strategies: prevention, treatment and recovery.

One prevention tactic is to pre-treat the beeswax to prevent wax moths from infesting bee hives. Burges and Bailey (1968) successfully used *Bacillus thuringiensis,* a gram positive bacterium found in soil, to pre-treat honeycomb which deterred wax moth infestation for up to two years without causing harm to the honey bees (Madigan et al., 2021)

While early efforts to use this technique were not practical due to the fact that *Bacillus thuringiensis* had to be mixed into the wax prior to use by the honey bees, advancements made in past decades allowed for formulations, notably, B401 and now B402 to be applied to

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empty honeycomb as a deterrent to wax moth infestation and comb damage.

It has been widely reported that the best way to prevent an infestation of *A. grisella* is to maintain a strong and healthy colony. Bees in a healthy colony will kill the *A. grisella* larvae and remove any web left behind (Solanki, et. al., 2020).

No threshold treatment level has been established for *A. grisella* (Hood, 2010). In 2020, a product called B402 (also known as Certan) was approved for use in killing wax moth larvae (Flemming, 2020). B402 uses *Bacillus thuringiensis* to selectively target and kill the wax moth while it is in the larval stage. The manufacturers, Vita Bee Health in collaboration with Valent Biosciences Corporation, claim B402 is up to 100% effective at killing wax moths in the larval stage (Flemming, 2020). Dr. Max Watkins, CEO of Vita Bee Health, claims that when used to kill wax moth larvae, B402 is harmless to the honey bees (Flemming, 2020). B402 is mixed with water and applied to combs prior to storage. The protection is said to last until the following season. No data regarding use or efficacy of B402 applied into a live colony were available (Flemming, 2020).

Wax moths traps may reduce the number of adult females outside the hive, but they are not effective for destroying larvae already inside the hive (Hood, 2010). The best treatment for control of *A. grisella* is to maintain a strong and healthy colony (Hood, 2010; Jack & Ellis, 2018; Solanki, et. al., 2020; *5 Ways to Eliminate Wax Moths in a Beehive PLUS Expert Prevention Tips*, 2021). However, sometimes maintaining a healthy hive is not enough to prevent wax moths from reproducing and feeding inside a hive as I will describe.

Figure 1. Hygienic uncapping behavior of honey bees suspected as a response to *A. grisella* larvae.

Identification of Bald Brood in a Healthy Colony

Bald brood due to the presence of *A. grisella* larvae was first described in 1942 by P. S. Milne of the Rothamsted Experimental Station (Milne, 1942). Milne described the phenomenon as one of bald-headed brood. He further describes observing "patches of uncapped cells containing apparently normal pupae, the glistening white heads of the latter giving the affected part of the comb a distinctly bald appearance." Milne comments that there were no signs of the usual brood diseases beekeepers were familiar with at the time. Additionally, when the pupae were removed from the comb, he found several "small black or dark-brown objects sticking to them" (Milne, 1942). These pellets were removed and observation under a microscope confirmed them to be feces from *A. grisella* larva. He also noted that some of the larva had small amounts of silk on them and an *A. grisella* larva was found to be living in one of the cells alongside a honey bee larva. The *A. grisella* larva was allowed to complete its life cycle and developed into an adult *A. grisella*, thereby confirming the diagnosis that "bald-headed brood" was caused by the presence of *A. grisella* larvae. In this article, I share evidence to support a similar finding based on hygienic bee behavior response and pathology of the affected frames.

Hygienic Bee Behavior

Uncapping and hygienic removal of larvae by honey bees as a response to brood diseases occurs over a wide variety of maladies such as American foulbrood (Park, 1937), chalkbrood (Guilliam, et al., 1983), *Varroa destructor* mites (Peng, et al., 1987) and small hive beetle (*Aethina tumida)* (SHB) larvae (Ellis, et al., 2003). This hygienic behavior in honey bees can be stimulated by freezing (Taber, 1982) or by piercing the larvae (Newton & Ostasiewski, 1986). The behavior is so common among honey bee colonies, that the uncapping and removal of larvae is used as the standard to measure the level of hygienic response exerted by individual colonies (Villegas & Villa, 2006). Hygienic behavior as a response to *A. grisella* is unique in that it occurs in patches or in lines based upon the route taken by the larvae within the comb, whereas hygienic behavior in response to high levels of *V. destructor* mites typically does not follow a pattern and is sometimes referred to as a pepperbox brood pattern because of the seemingly random pattern of uncapping larval cells (Department of Primary Industries and Regional Development et al., 1993; Bee Informed Partnership, 2013; Shimanuki & Agricultural Research Service, 1991).

A Case of Bald Brood Caused by *A. grisella* Larva in a Healthy Hive

Figure 1 shows a linear pattern of uncapping cells containing larvae. The

Figure 2. Honey bee larvae containing purported *A. grisella* feces.


Figure 3. Larvae removed from cells of an infected hive.


Figure 4. Uncapped larva from hygienic behavior.


Figure 5. Small hive beetle larvae on a brood frame (Martin, n.d.).

pattern is linear and suggests it was done in response to *A. grisella* larvae.

Additionally, Milne (1942) described small black or dark-brown objects sticking to the larvae. The larva removed from the uncapped cells had small black or dark-brown objects adhering to their distal end (Fig. 2). The characteristics of the small black or dark-brown objects suggest they are the feces of *A. grisella* larva. In some instances, a similar condition occurs with American foulbrood, but the remains of larvae in colonies infected with AFB become dark brown and glue-like (de Graaf et al., 2006), and do not remain white like the larva I observed. Evidence also does not support chalkbrood, a fungal disease caused by *Ascosphaera apis*, as a cause. Larva infected with *A. apis* will die inside their capped cell (Bee Aware, 2014). Worker bees remove the cap and expose the dead larvae which will have a white, mummified, chalky appearance (Bee Aware, 2014). The larvae I observed remained white, intact and solid without a chalky appearance (Fig. 3). When worker bees uncap larvae in colonies infected with *V. destructor* mites, the worker bees chew the larva from the head down to remove it from the cell (Oliver, 2019), but the larva I observed in this colony remained largely intact (Fig. 4). In fact, only a small number of larvae in any of the photos showed evidence of being chewed by worker bees. Finally, while there was one small hive beetle present on the inner cover of the hive, an infestation of small hive beetles is characterized by slimy, discolored comb as well and masses of beetle larva on the frame (Fig. 5, Torgerson et al., 2016). There is no evidence of either slimy comb or larvae in any of the figures.

History, Treatment and Mitigation of This Hive

This hive was started, along with two others on May 23, 2021, using five frames of brood and a New World Carniolan Queen in a queen cage with a sugar plug. An inspection on June 8, 2021 revealed that the queen had been released and new eggs were present in the brood box. By June 29, 2021 the colony had combed eight of the frames in the 10-frame brood box and a second 10-frame brood box was added. On July 15, 2021, a full treatment of two strips of Mite Away Quick Strips (MAQS) were placed between the brood boxes. The weather unexpectedly turned warm in the next few days with high temperatures reaching 91°F on July 17 and July 18, 2021. This likely was a very stressful event for the colony as the manufacturers of MAQS note in their package insert that temperatures above 85°F can be harmful to the honey bees especially during the first three days of treatment (Application MAQS USA – NOD Apiary Products, n.d.). To support this suspicion, very few eggs were noted on the July 30, 2021 inspection and no eggs or sign of the queen were found on the August 5, 2021 inspection. The queen or eggs were present in the other two hives in the apiary on August 5, 2021. The queen was presumed to be dead in the colony of discussion so a new New World Carniolan was introduced into the colony inside of a queen cage with a sugar plug on August 10, 2021. Nine days later during an inspection on August 19, 2021, new eggs were found in the top brood box. A mite check using an alcohol wash yielded two mites so a full treatment of two strips of MAQS were placed between the brood boxes of the hive. Nothing remarkable was noted on the weekly inspection notes until the discovery of what evidence suggests is bald brood was noted on the September 28, 2021 inspection. The colony was fed a mixture of 1:1 sugar/water weekly beginning July 8, 2021 through September 2, 2021, after which they were fed a mixture of 2:1 sugar/water until October 28, 2021. A treatment of oxalic acid vaporization was administered on November 28, 2021. It needs to be noted that feeding the bees during the mite treatment with MAQS may have been stressful to the honey bee colony as current treatment guidelines for using MAQS advise to not feed honey bees during the treatment period (The Honey Bee Health Coalition & Caron, 2018).

No treatment was administered to the hive in this case study other than removing the uncapped larvae to try to diagnose the malady. The frames were returned to the hive and the colony was able to remove any *A. grisella* larvae present. The hive survived the 2021 – 2022 Winter in Southwest Connecticut and as of August 15, 2022, this hive is alive and thriving. It yielded 48 pounds of honey harvested July 5, 2022.

My notes regarding this hive reveal that it was healthy and unremarkable for at least six weeks prior to discovering bald brood. Both brood boxes were full of bees and there was no empty space noted. By October 7, 2021, the entire top brood box was full of honey and the brood nest had moved to the lower brood box. This was only one week after the discovery of the bald brood. I believe this speaks positively to the healthy state of the hive in both the time period leading to the emergence of the bald brood, as well as the time period following the discovery. It is for this reason that this case is unusual and it is why I decided to share what I learned.

Advice for Beekeepers

Because there are no effective treatment measures for *A. grisella*, prevention tactics are the best approach for managing this hive pest. A few tactics to prevent *A. grisella* from becoming a problem within a hive are listed below (*5 Ways to Eliminate Wax Moths in a Beehive PLUS Expert Prevention Tips*, 2021; Hood, 2010; Jack & Ellis, 2018; Solanki, et al., 2020):

Keep your bee colony strong and healthy. As unhealthy colonies begin to decline, the number of worker bees available to keep the hive clean declines. Pests that feed on the waste and other products of bees begin to establish a presence, which can include *A. grisella.* Bees in a healthy colony will kill *A. grisella* larvae and remove any web left behind.

Keep a low bee to comb ratio. Healthy colonies that experience frequent swarms lose large numbers of bees in the process. Adjusting the number of brood boxes to fit the number of bees can help maintain a low bee to comb ratio making it more likely that worker bees will be able to service and clean all of the comb in the hive. Additionally, bees may come out of Winter with only a small number of bees. Careful Spring management of hive space will allow for the remaining bees to more easily find and remove pests that may be present in the comb.

Raise hygienic bees. Bees can be selected to be more resistant to certain pests. Bees that have been bred to be resistant to *A. grisella* and other pests tend to be better housekeepers, meaning they exhibit more frequent or strong behaviors associated with keeping the hive clean and free of pests.

Replace old comb with new foundation. Adult female *A. grisella* prefer to lay their eggs in darker, older comb. Even though drawn comb has been referred to as "a beekeeper's most valuable possession" (Imirie, n.d.), it is important to replace comb that presents an ideal egg laying site for *A. grisella.* Beekeepers should not be afraid to discard older, darker comb.

Use traps to trap adult females outside the hive before they can lay their eggs. There are currently no commercial *A. grisella* traps for sale in the United States. However, there are several homemade traps which can be easily made from readily available, low cost materials such as water, sugar, vinegar and banana peels in a two liter bottle. An internet search for "Wax moth traps" will yield several versions of this trap.

*(#**6** from Jerry… Apply B402 to empty comb as a biocontrol for Wax Moths)*

Conclusion

Grant Gillard (2009) summed up the bane of *A. grisella* nicely when he said, "Wax moths keep us from becoming lazy. Conversely, they make us pay dearly for our procrastination. They wake us up from lethargy and reinforce our resolve in how we have to be better beekeepers and more efficient managers of our resources." BC

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