

BIOAg Project Report

Report Type:

Progress

Title:

Investigating the effects of microbial biopesticides as biocontrol for honey bee Varroa mites

Principal Investigator(s) and Cooperator(s):

Brandon Hopkins

Abstract:

Metarhizium is a sporulating soil fungus used worldwide as pest biocontrol agents for its entomopathogenic properties. Varroa mite is a major pest of honey bees and is detrimental to honey bee health and is the primary cause of colony collapse. Chemical miticides raise concerns about human and pollinator health hazards and adverse environmental impacts. Therefore, the beekeeping community actively seeks biopesticides as an alternative solution. This research proposal aims to investigate the potential of a *Metarhizium* isolate (*Metarhizium brunneum*), as a biopesticide to control Varroa mites in honey bee colonies. The use of *Metarhizium brunneum* for Varroa mite control is experimental and not currently a labeled use. We also aim to select new strains of *Metarhizium brunneum*, more tolerant to higher hive temperature and can effectively kill Varroa mites. Our study will also provide crucial information on how this microbial biopesticide might improve directly honey bee health. This project aligns with the BioAg priority of advancing applied research of sustainable management of honey bees pests and provide cost-effective and eco-friendly biocontrol strategies.

Project Description:

In this project we seek to investigate the potential of a *Metarhizium* isolate (*Metarhizium brunneum*), as a biopesticide to control Varroa mites in honey bee colonies in large scale field trials over time. We also aim to select new strains of *Metarhizium brunneum*, which are more tolerant to higher hive temperature and can effectively kill Varroa mites. This project also aims toward advancing applied research of sustainable management of honey bees pests and provide cost-effective and eco-friendly biocontrol strategies.

Objectives

- (1) Assess whether current *Metarhizium* biopesticide controls Varroa mites numbers in honey bee colonies over time
- (2) Test if *Metarhizium* treatment also improve honey bee health
- (3) Select new strains of *M. brunneum* that will be more tolerant to higher hive temperature and more virulent to Varroa mites

Outputs:

Overview of Work Completed and in Progress:

Initial field trials conducted in spring 2024 to test the efficacy of *Metarhizium brunneum* failed due to ineffective application method. The trial was repeated on a smaller scale with a revised application method in fall 2024 and returned promising results. Samples of bees have been collected and are

awaiting PCR analysis to determine if *Metarhizium* treatment improved honey bee health. Cultures of *M. brunneum*, along with other entomopathogenic fungal strains, are currently being developed to withstand higher temperatures and those virulence bioassays will begin in early spring 2025 to determine efficacy.

Methods and Results:

Methods—The spring 2024 trial was conducted using 48 honey bee colonies in two different apiaries. Control hives had a brown paper bag of brown rice (N=24) and treatment hives had a brown paper bag with *M. brunneum* culture on brown rice with spores (N=24). *M. brunneum* was initially cultured on agar and then moved to brown rice to mass produce spores in brown paper bags. Bags were placed on the top of each hive to allow bees to gradually remove each grain of rice while passively spreading spores throughout the hive. Mite washes (100 nurse bees shaken in 70% ethanol) were done biweekly for two consecutive months to monitor the mite load within the colony. This method failed to yield results observed in the laboratory and was therefore revised. The revision required the separation of spores from the rice using a 0.01% Tween 80 solution and then directly spraying the spore solution onto each side of every frame in the hive. Due to the time and resource loss with the first trial, the revised trial included eight hives, *M. brunneum* spore solution treated hives (N=4), and 0.01% Tween 80 solution control hives (N=4) in one apiary. The initial mite counts were recorded prior to beginning study via mite wash. Daily mite drop was recorded every 2-4 days using sticky cards placed at the bottom of the hive. Nurse-bee samples were collected prior to *M. brunneum* application and were collected weekly for a total of four weeks. These samples will be used to determine if *M. brunneum* indirectly improved honey bee health by reducing disease vectoring *Varroa destructor* mites from the colony. The development of more heat tolerant fungal strains is being conducted in incubators under high humidity conditions to replicate the internal hive environment. Strains that produce growth will be selected and cultured to conduct virulence trials in the early spring 2025.

Results—The spring 2024 trial produced no significant differences in mite control when compared to control hives that did not receive mite treatment. The fall 2024 trial showed treatment and control colonies had no significant difference in initial mite populations (Figure 1). After bootstrapping and managing missing data (due to small sample size and colony loss), a Kruskal-Wallis test with repeated measures showed significantly higher mite drops on day 4 ($p = 0.0339$, $H = 4.5$) and day 15 ($p = 0.0209$, $H = 5.33$) in *M. brunneum* treated colonies when compared to daily mite drop in control colonies (Figure 2). There was an overall significant difference between treatment and control, where the treated colonies dropped more mites than the control colonies ($p = 0.04306$, $H = 247$), thus providing evidence that *M. brunneum* may be an effective IPM control tool for *Varroa destructor* mites in honey bee colonies.

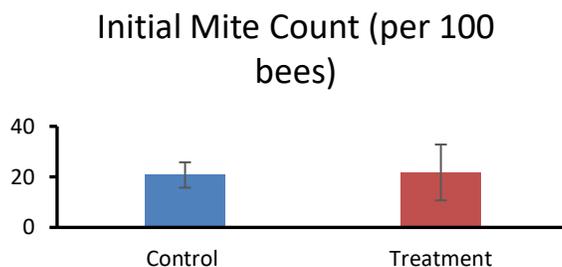


Figure 1. Initial mite counts per 100 bees.

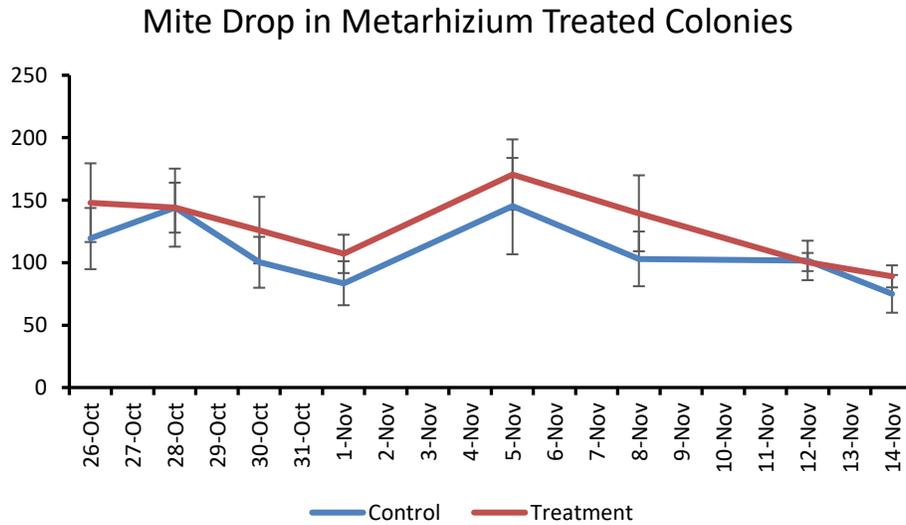


Figure 2. Mite drop in control vs treated colonies, fall 2024.

Publications, Handouts, Other Text & Web Products:

Expected Summer 2025

Outreach & Education Activities:

Beekeepers in the Pacific Northwest have been notified of our work and results via presentations, posters, and conversations during beekeeper meetings. Beekeepers are very excited about our work and eagerly await the conclusion of our studies with hopes to incorporate entomopathogenic fungi in their future mite control programs.

Impacts:

Short-Term:

Preliminary results show *M. brunneum* is a promising biological control for *Varroa destructor* mite management in honey bee colonies.

Intermediate-Term:

Through conversations and presentations, beekeepers are learning the importance of integrated pest management and the benefits of biological control tools when managing mite populations, while also preventing the development of mite resistance to heavily used miticides.

Long-Term:

Evidence of *M. brunneum* efficacy with revised application methods applied inside honey bee colonies to control mites is laying the ground work for companies to begin developing biocontrol products for mite management in honey bee colonies.

Additional Funding Applied for/Secured:

2025 Specialty Crop Block Grant, \$250,000, Titled: Extension implementation of integrated pest management in honey bee colonies

A minimum of two more grants will be applied for prior to the conclusion of this grant.

Graduate Students Funded:

Molly Quade, Master's of Science, Entomology Department

Recommendations for Future Research:

Future research should focus on the application of entomopathogenic fungi with a focus on spore dispersal targeting the brood area within a colony.