POTENTIAL USE OF ENTOMOPHATOGENIC FUNGI AGAINST *Varroa destructor* (Acari: Varroidae).

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Introduction

An alternative to reduce damages caused by *Varroa destructor* is the biological control with entomophatogenic fungi. The Chilean Institute of Agricultural Research, INIA, has a collection of native entomophatogenic micro-organisms which includes 800 isolates of entomophatogenic fungi *Beauveria bassiana* and *Metarhizium anisopliae*. Consequently, the objective of this work was to select, through laboratory and field trials the most virulent and specific isolates to develop a biological acaricide for management of *V. destructor* on hives.

Entomophatogenic Fungi Collection

Beauveria bassiana

Metarhizium anisopliae

Materials and Methods

Laboratory Trials

40 *M. anisopliae* and 42 *B. bassiana* isolates were evaluated at different temperatures (15-35°C) to select resistant isolates to survive at honey bee hives temperature (30-35°C).

The selected isolates were inoculated by spraying a suspension of 10^7 conidia mL^{-1} directly on adult of *V. destructor* through a Potter tower. The treated mites were maintained on honey bee pupae. Mortality of *V. destructor* with different isolates were registered daily and the dead mites were incubated in wet chamber.

The best isolate "Qu-M845 *M. anisopliae"* was evaluated at different concentrations (10^5, 10^6, 10^7, and 10^8 conidia mL^{-1}) on *V. destructor* adults to determine the Lethal Concentration 50 (LC50) in laboratory conditions.

Mites before inoculation

Mites after inoculation

Field Trials

A field trial was performed through three different application methods: T1. Filter paper imbibed with conidia located inside the hive, T2. Dry conidia were powdered on and between the frame hive, and T3. Conidia dispenser at the hive entry. The control test (T0) was a hive without fungi. The dose for each method was $5x10^5$ conidia for beehives, and applied on early fall. Level infestation of *V. destructor* was estimated before and after applications, and mites mortality per day was detected using a sticky cardboard located on the bee hive floor.

Conidia on filter paper (T1)

Dry conidia (T2)

Conidia Dispenser (T3)

Results

Pathogenesis differences were founded among isolates (Tukey, $P \leq 0.05$) in the screening test.

The most pathogenic isolates of *M. anisopliae* were Qu-M845 and Qu-M326 which caused the highest rates of mortality of *V. destructor*. *B. bassiana* did not produce significant mortality (Figure 1) to the pest.

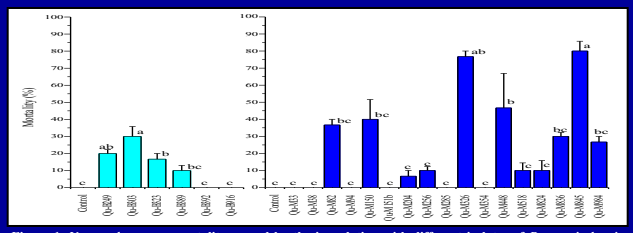


Figure 1. ***Varroa destructor*** **mortality caused by the inoculation with different isolates of** ***Beauveria bassiana*** **(Qu-B) and** ***Metarhizium anisopliae*** **var.** ***anisopliae*** **(Qu-M).**

Qu-M845 caused 98% of mites mortality 7 days post inoculation (dpi), with 10^8 conidia mL^{-1}, which was statistically similar ($P \leq 0,05$) with 10^7 conidia mL^{-1} (72% of mortality). The LC50 and LC90 for this isolate were about $10^{5.41}$ and $10^{7.59}$ conidia mL-1, respectively (Figures 2 and 3).

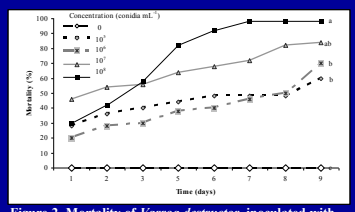


Figure 2. Mortality of ***Varroa destructor*****, inoculated with different concentrations of** ***Metarhizium anisopliae*** **var.** ***anisopliae*** **Qu-M845 isolate.**

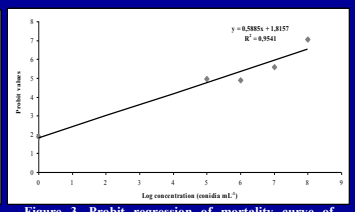


Figure 3. Probit regression of mortality curve of ***Varroa destructor*****, inoculated with different concentrations of** ***Metarhizium anisopliae*** **var.** ***anisopliae*** **(Qu-M845) isolate.**

The level of infestation of mites before treatment ranged between 5.41 to 7.42 % in the hives. After application all *M. anisopliae* treatments showed lower mite infestation level than the control which increased in 71.143%. The best treatment was dry conidia powdered in the hive which reduced in 67.03% the level of infestation with *V. destructor* (Figure 4).

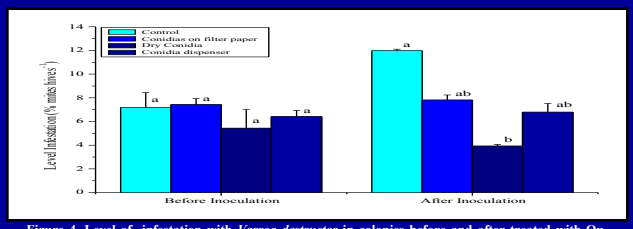


Figure 4. Level of infestation with ***Varroa destructor*** **in colonies before and after treated with Qu-M845 isolates of** ***Metarhizium anisopliae*** **var.** ***anisopliae*****.**

Cumulative mortality of *V. destructor was s*ignificantly different ($P \leq 0.05$) between bee colonies 21 dpi. At the same time, the cumulative daily mortality of the honey bees did not differ significantly ($P = 0.002$) between treatments (Figures 5 and 6).

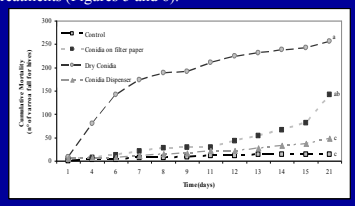


Figure 5. Cumulative mortality of ***V. destructor*** **over time with different methods of application of** ***Metarhizium anisopliae*** **var.** ***anisopliae*** **Qu-M845 isolates.**

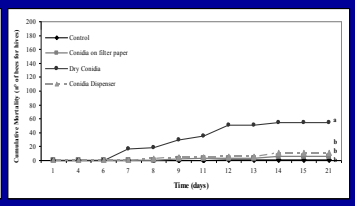
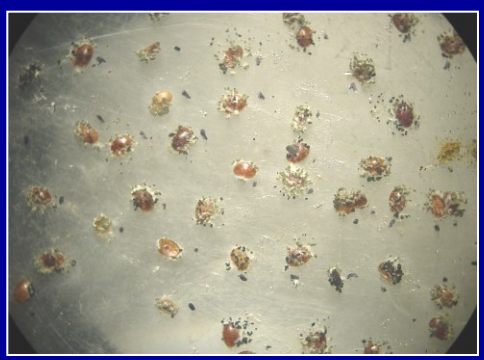


Figure 6. Cumulative mortality of honey bees over time with different methods of application of ***Metarhizium anisopliae*** **var.** ***anisopliae*** **Qu-M845 isolates.**



Metarhizium anisopliae **var.** ***anisopliae*** **on** ***Varroa destructor*****.**