

# Sampling and evaluation of entomopathogenic fungi for the control of the two-spotted stink bug

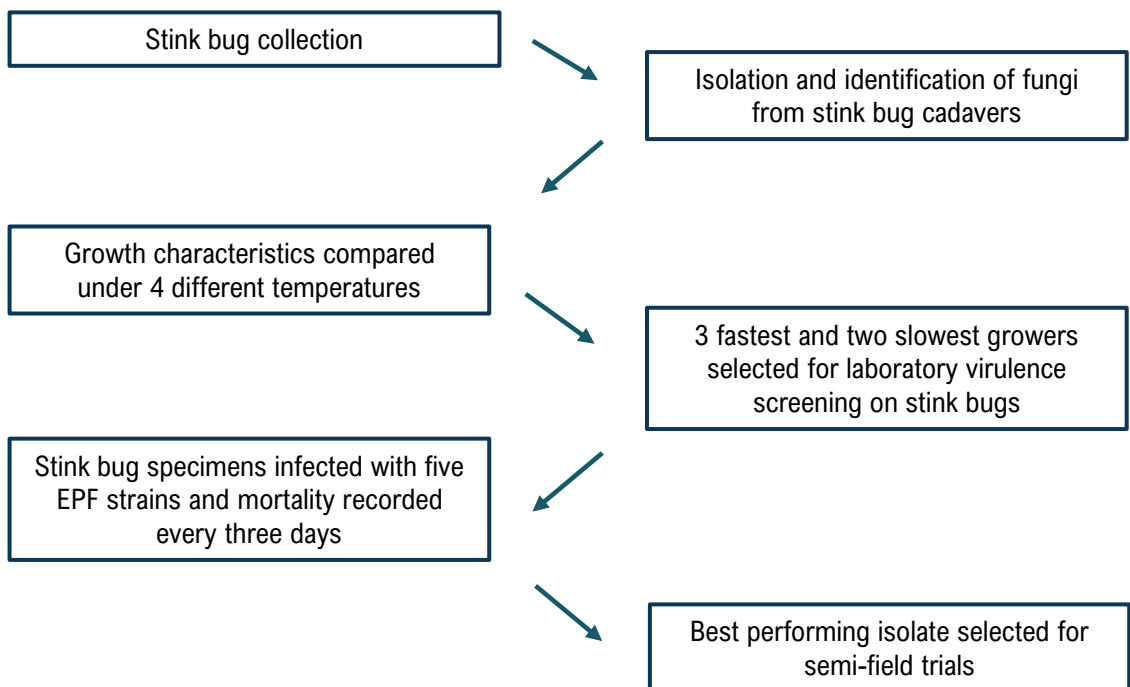
Strains isolated from stink bugs significantly outperformed the commercial product

## Problem

- The two-spotted stink bug is the most abundant and damaging stink bug species in South African macadamia orchards.
- Entomopathogenic fungi (EPFs) are fungi which infect and kill insects, and finding a species or strain adapted to stink bugs may improve the efficacy of these mycoinsecticides.
- EPFs are an important control option in the macadamia IPM toolbox as insecticides are increasingly coming under threat as a result of legislative (health and environmental risks) and resistance issues.

## Aims

- To isolate EPFs from stink bugs which are highly virulent (harmful, causing the death of the stink bug) to stink bugs and endemic to macadamia-growing regions.



### Semi-field trials

Two-year old 695 seedlings placed in foldable insect cages on the south-facing side under mature macadamia trees. Seedlings sprayed with either the experimental *Beauveria bassiana* isolate, the commercial *Beauveria bassiana* product, cypermethrin or acephate.

Immediately after spraying, five newly-emerged two-spotted stink bug specimens placed in each cage, and stink bug mortality assessed every three days. Mortality due to fungal infection recorded by allowing for fungi to sporulate.

## Results

- Thirteen groups of fungi were isolated and identified from stink bug cadavers, of which two were known to act as EPFs (*Beauveria bassiana* and *Purpureocillium lilacinum*).
- 11 isolates of *B. bassiana* isolated from two-spotted stink bug specimens and a green vegetable bug were subjected to growth trials to identify five isolates for laboratory virulence trials (ability of an organism to infect the host and cause a disease or death) on two-spotted stink bug specimens. The three fastest and two slowest growers were selected to account for any variation in sporulation, virulence etc.
- Lab-reared two-spotted stink bug specimens were inoculated with the five *B. bassiana* isolates, and mortality was assessed every three days. Mortality due to fungal infection was recorded and confirmed by allowing fungi to sporulate.
- Mortalities increased with time after application of *B. bassiana* isolates. The isolate of *B. bassiana* which performed best in the laboratory experiment (50% mortality at 6.99 days and 90% mortality at 12.85 days) was subjected to a semi-field trial. The experimental EPF was also compared with a commercially available *B. bassiana* product, as well as cypermethrin and acephate.
- At most time points post-inoculation, the EPF strains isolated from stink bugs significantly outperformed the commercial product in terms of percentage mortality. The percentage mortality was between 13% and 35% higher at different days post-inoculation.
- The insecticides cypermethrin and acephate had a faster knock-down of stink bugs, but after 12 to 18 days post-inoculation there was no significant difference between the endemic EPF and the chemical insecticides in terms of percentage mortality.



**Stink bug cadavers with EPF sporulating after killing the specimen.**

## What does this mean?

- The isolate of *B. bassiana* isolated from stink bugs performed better than the commercially available product.
- Although chemical insecticides had a quicker knock-down effect, the EPFs were able to achieve the same level of control 12 to 18 days after inoculation.
- Future work includes optimizing factors such as UV sensitivity, registration trials and compatibility tests with registered insecticides for an IPM strategy.



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# Evaluation of potential EPFs as biocontrol options against stink bugs

## Problem

- Biological control options for stink bugs, such as entomopathogenic fungi are important tools in the macadamia industry's IPM toolbox. *Beauveria bassiana* and *Metarhizium anisopliae* are registered for use on macadamias and are widely used.
- Other groups of fungi besides *Beauveria bassiana* have been isolated from dead stink bugs and soil, and their potential value as alternative native EPF's (stink bug-specific strains of EPF) are unknown.

## Aims

- To identify the fungal groups isolated from dead stink bugs and soil based on DNA sequences.
- To determine the potential of these fungi as EPFs. Lab-reared stink bugs were inoculated with these fungi, and mortality of stink bugs were recorded.

## Results

- The following fungal groups were identified: *Aspergillus*, *Fusarium*, *Bionectria*, *Purpureocillium*, *Chaetomium*, *Neopestalotiopsis*, *Pestalotiopsis*, *Myriodontium* and *Scopulariopsis*.
- The *Aspergillus*, *Fusarium*, *Bionectria*, *Purpureocillium*, *Chaetomium* and *Scopulariopsis* groups of fungi contain known EPF species, and species from these groups were tested on stink bugs.
- None of these fungi performed better than a native *Beauveria bassiana* isolate currently undergoing trials at the ARC.

## What does this mean?

- These fungal groups are secondary fungi and should not be included in further studies.
- The four commercial products were all effective against a lab-reared stink bug population.

