### Exploration of biological control strategies for the polyphagous shot-hole borer





AGRISCIENCES EYENZULULWAZI NGEZOLIMO **AGRIW/ETENSKAPPE** 

### Dr. Steffan Hansen

## **Research** Career

- 1/25.
  - 2020-2023 -> PhD, first post-doc at Stellenbosch University: Research into the **\*taxonomy**, pest status, biological control and physiology of a South African weevil pest
  - \*Taxonomy part of a very successful collaboration with CIRAD, INRAE at CBGP

Continuing with many more weevils to describe!







## Research Career

2/25.

In 2022, polyphagous shot-hole borer (**PSHB**) attacks **deciduous** fruit (pear) for first time worldwide in SA.

• Pecan nuts also attacked, with potential risk to macadamia\*

Oct 2023 -> I obtain funding from industry bodies (Hortgro Science, SAPPA, SAMAC) for a three year postdoc at SU





# Background

- 3/25.
  - Euwallacea fornicatus (PSHB) is a Scolytinae ambrosia beetle native to South-East Asia
    - Vectors **ambrosia fungi** into tree, upon which it and its progeny **feeds**
  - In past 20 years, it starts invading California, Israel, South Africa (Paap et al., 2018). Damage seen on avocado, urban and native trees

• Closely associated ambrosia fungus (*Fusarium euwallacaea*) described by Freeman et al. (2013)







# Beetle & Fungus

4/25.









# Damage and Symptoms

5/25. One of very few ambrosia beetles to attack living trees

• Causes Fusarium dieback (FD) in more than 100 species of host tree -> new susceptible hosts get added to list

• FD -> Mechanical damage by beetle (girdling), blocking xylem by *F. euwallaceae* 

• Kills very susceptible hosts (e.g. *Acer negundo*) in a few months





### PSHB and FD













# Other PP Fungi

7/25.

Other plant pathogenic fungi (Graphium euwallaceae; *Paracremonium pembeum*, etc.) found in beetle mycangia and galleries (Lynch et al., 2016)



• Various opportunistic wood-rot/canker causing pathogens -> on beetle integument and/or infect after damage is caused?. e.g. Phaeacremonium minimum







# Economic Impact

8/25. Can cause extreme damage to native ecosystems, and urban trees

• Widespread tree death not reported for cultivated crops, but has the potential to cause serious economic harm

Cost to SA estimated to be multi-billion USD (tree loss, removal, etc. De Witt *et al.*, 2022)

Attacks avocado, pears, apples, plums, pecan nut, macadamia nut





# **Control Options**

9/25. Monitoring

> • Removal of excellent reproductive hosts (A. negundo, Quercus *robur* ) where feasible

• Removal and sanitation of infested plant material

Chemical injection -> Limited efficacy, potential for harm to tree



# Control Options

10/25.

# Beetle and fungus deep in gallery -> Difficult to reach



![](_page_10_Picture_5.jpeg)

![](_page_10_Picture_6.jpeg)

![](_page_11_Picture_0.jpeg)

# The Role of Mites...

11/25.

- Multiple mite species associated with bark-and-ambrosia beetles
- Ecological roles: detritivores, fungivores, predators, parasitoids

Some live on bark, some inside galleries, many are **phoretic** (i.e. use beetles as transport)

![](_page_11_Picture_6.jpeg)

![](_page_11_Picture_7.jpeg)

![](_page_12_Picture_0.jpeg)

## The Role of Mites...

12/25.

![](_page_12_Picture_3.jpeg)

![](_page_12_Picture_4.jpeg)

Hofstetter et al., 2015

![](_page_12_Picture_6.jpeg)

![](_page_12_Picture_8.jpeg)

![](_page_13_Picture_0.jpeg)

### 13/25.

## The Role of Mites...

![](_page_13_Picture_4.jpeg)

![](_page_13_Picture_5.jpeg)

![](_page_14_Picture_0.jpeg)

# The Role of Mites...

14/25.

Can we use fungus feeding mites to vector Microbial Biocontrol Agents to the galleries? (Berto et al., 2022).

- Mites can act as **dispersers** of **fungal** propagules
  - Specialized relationships with mutalists (e.g. Tarsonemus spp. in Dendroctonus frontalis (Curculionidae: Scolytinae) galleries
  - Less specialized, on cuticle or through ingestion/excretion (Astigmata)

![](_page_14_Picture_7.jpeg)

![](_page_14_Picture_9.jpeg)

### Hoftstetter & Moser, 2014

## Collection

### 15/25.

- We collected mites from:
  - Trapping beetles in field (quicevorol baited dry funnel traps)
  - Collecting infested material -> Emergence bucket in lab
  - -> 1) Collect from emerging beetles, 2) surface of logs near galleries, 3) splitting open logs

• Cultivated (pecan) and ornamental material from SA, also collection from Vietnamense samples (FABI quarantine facility)

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![](_page_15_Picture_8.jpeg)

![](_page_16_Picture_0.jpeg)

- 16/25.
  - We've collected 15+ species of mites associated with PSHB (and some other bark beetles) from SA (invasive) and 5 + species from Vietnam samples -> ID
    - Morphological (relevant experts)
    - Molecular (?) Spider mite direct PCR protocol? (Sakamotoh & Gotoh, 2017)

Mainly Mesostigmata, Astigmata, some Tarsonemidae

![](_page_16_Picture_6.jpeg)

![](_page_16_Picture_10.jpeg)

### Culture

- 17/25.
- 5 species from SA currently in culture
  - Feed and reproduce on **autoclaved barley** inoculated with F. euwallaceae in sterile 50 ml centrifuge tubes
  - Three Astigmatan species (potentially Acaridae), two of which form hypopi that are phoretic on beetles
  - Two Uropodine mesostigmata (Uropodina?), one established phoretic on E. fornicatus, one collected in galleries of a Platypus spp.

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![](_page_17_Picture_7.jpeg)

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# Going Forward...

18/25.

Test ability of these mites to feed and reproduce on biocontrol fungi (such as *Beauveria*, *Trichoderma*)

• Grow fungi on cellophane on growth media, remove cellophane and add 5 mites -> Assess for survival, feeding and reproduction

Assess whether mites can vector viable fungal propagules on cuticle, and intestine (surface sterilization) on sterile growth media

![](_page_18_Picture_6.jpeg)

![](_page_18_Picture_7.jpeg)

![](_page_19_Picture_0.jpeg)

# Going Forward...

### 19/25.

Assess whether mites can vector biocontrol fungi into active E. *fornicatus* galleries, and assess for control efficacy, in laboratory colonies.

Start thinking about an in-field delivery mechanism....

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![](_page_19_Picture_7.jpeg)

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# What will the mites potentially vector??

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![](_page_20_Picture_4.jpeg)

![](_page_20_Picture_6.jpeg)

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# Microbial Biological Control

### 21/25.

Entomopathogenic fungi, e.g. *Beauveria* -> Attack beetles 

Antagonistic fungi, e.g. *Trichoderma* -> Attack or outcompete plant pathogenic fungi

• Lots of local isolates, as well as commercial products for general biological control.

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![](_page_21_Picture_11.jpeg)

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# Entomopathogenic Fungi

### 22/25.

Some commercial isolates tested (Nel et al., 2023) – Works well in petri-dish, but not in infested plant material

• Colonies of *E. fornicatus* in emergence cages often killed by Beauveria (e.g. Beauveria bassiana)

• Two native, one commercial isolate of *B*. bassiana to be screened on *E. fornicatus* females allowed to establish colonies in growth media in tubes

![](_page_22_Picture_10.jpeg)

![](_page_22_Picture_11.jpeg)

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# Antagonistic Fungi

23/25.

- Trichoderma atroviridae : Fast growing fungus that shows antagonistic effects against other fungi
  - Some isolates used for **bio-control of wood pathogens**, applied on pruning wounds
- Three isolates tested easily outcompetes F. euwallaceae, other mycangial fungi and wood-rot pathogens *in vitro* (preliminary results).
- How does *T. atroviridae* and *Beauveria* spp. interact?

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## Future Work

24/25.

- What role do **phoretic nematodes** associated with PSHB play?
- Commonly found in galleries

- Two bacterial feeders, one fungal feeding species -> pending identification
  - Can we potentially exploit these as well?

![](_page_24_Picture_7.jpeg)

![](_page_24_Picture_10.jpeg)

![](_page_25_Picture_0.jpeg)

### Team

25/25. Dr. Steffan Hansen (Postdoctoral Fellow, SU)

- Prof Francois Roets (Host, SU)
- Dr. Davina Saccaggi (Collaborator, CRI, SU)
- Prof Edward Ueckermann (Collaborator, NWU)
- Prof Lizel Mostert (Collaborator, Plant Pathology, SU)
- Ms. Annelie Smit (Research Assistant, SU)  $\bullet$
- Hortgro Science, SAPPA, SAMAC, BlueBug ID service industry partners

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• Hortgro Science, SAPPA, SAMAC for funding

• FABI quarantine facility (B. Hurley, A. Collop)

• P. Barker (arborist, Capetown)

SU nematology (A. Malan, C. Kapp)

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![](_page_26_Picture_10.jpeg)

Macadamias South Africa NPC

![](_page_26_Picture_12.jpeg)

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## Comments/Questions?

• Thank you for your attention!

• For any questions or comments on this work, please feel free to contact me on shansen.rsa@gmail.com

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![](_page_27_Picture_5.jpeg)