Occurrence of *Botrytis cinerea* across honeybees, hives, and blueberry flowers and fruit on farms in the Western Cape, South Africa

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1 Introduction

Pollination by honeybees (*Apis mellifera*) is an essential ecological service, particularly in South Africa's growing blueberry industry. Honeybees may, however, also act as vectors of *Botrytis cinerea*, a destructive fungal pathogen causing blossom blight and grey mould of blueberries. Disease caused by *Botrytis* significantly impacts fruit quality and quantity.

2 Aim

To investigate the presence of *B. cinerea* on honeybees and hives in Western Cape blueberry farms, and determine whether its presence is related to disease incidence on blueberry flowers and fruit

Objectives

- Determine the presence of *B. cinerea* on honeybee workers and hives in Western Cape blueberry farms
- Determine whether *B. cinerea* incidence on honeybees and hives is related to incidence on blueberry flowers and fruit

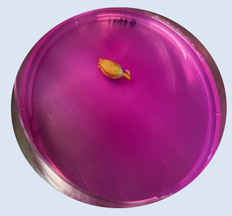
3 Methods

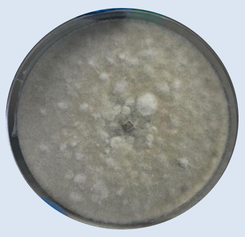
a. Sample collection

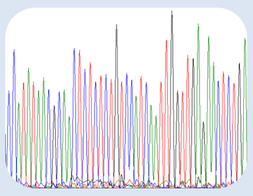
- Six commercial blueberry farms in the Western Cape
- Two different blueberry varieties
- Farms categorised into historic *B. cinerea disease* pressure (high, medium and low)
- Samples collected:
  - Blueberry fruits
  - Blueberry flowers
  - Hive entrance swabs
  - Honeybee workers

c. Statistical analysis

- R and RStudio
- Generalized linear fixed-effects models (binomial data)
- Linear regression







b. Sample processing

Samples plated on *Botrytis* selective media
↓
Plates incubated at 25 °C in the dark
↓
Plates grouped based on morphology
↓
Subcultured + hyphal tipped
↓
Pure *B. cinerea* cultures
↓
DNA extracted
↓
PCR + gel electrophoresis
↓
Sanger sequencing

4 Results

***Botrytis cinerea* presence on flowers and blueberries:**

- Three-way and two-way interactions
- Main effects:
  - Status
  - Variety

} Not significant ($p > 0.05$)

***Botrytis cinerea* presence on honeybees:**

- Significantly lower on medium pressure compared to high pressure farms ($p < 0.05$)
- Two-way interaction
- Main effects:
  - Status
  - Variety

} Not significant ($p > 0.05$)

***Botrytis cinerea* presence on hive swabs:**

- Main effects:
  - Variety

} Not significant ($p > 0.05$)

  - Status

Regression:

- Positive slope
- Relationship not significant

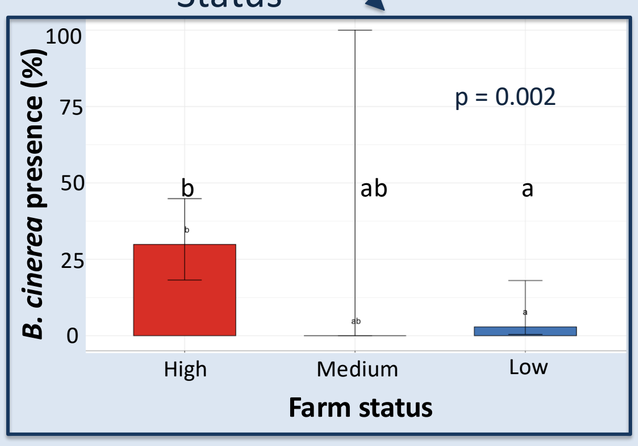


Figure 1. Effect of farm status on *Botrytis cinerea* presence on hive entrance swabs

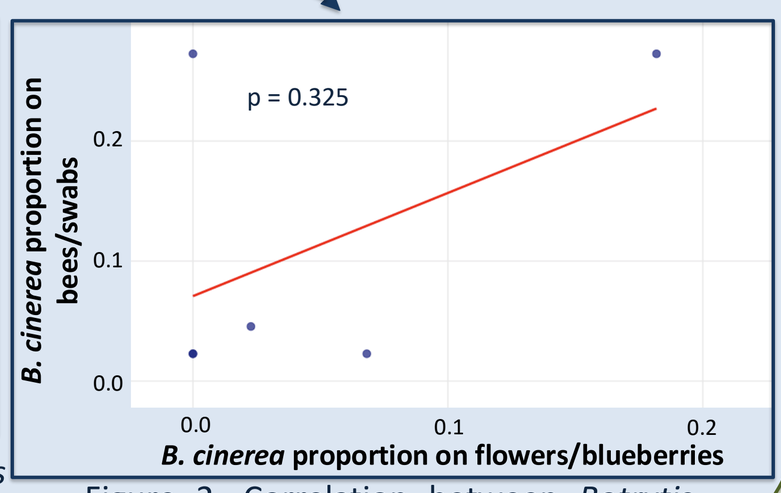


Figure 2. Correlation between *Botrytis cinerea* on blueberry plants and honeybees/swabs

5 Discussion and Conclusion

- *Botrytis cinerea* was isolated from blueberry flowers, fruits, honeybees and hive entrances
- In this study, the presence of *B. cinerea* on:
  - Blueberry flowers, fruits and honeybees was not significantly predicted by Farm Status or Variety
  - Honeybee hive entrances was significantly predicted by historically observed *B. cinerea*
    - *Botrytis cinerea* is ubiquitous and produces overwintering structures that survive on plant debris and on the soil surface
    - The highest amount of *B. cinerea* was isolated from honeybee hives placed near the soil surface
- No significant relationship was found between the incidence of *B. cinerea* on blueberry plants and on the honeybees and hive swabs
- Honeybees are not a primary source of inoculum spread under the tested conditions
- If a farm has a history of high *B. cinerea* presence, control methods should be put in place to reduce inoculum buildup
- The control of *B. cinerea* is difficult, and environmental factors should be taken into consideration when planning management strategies

References available upon request