

# Monitoring fungicide sensitivity of the powdery mildew and Colletotrichum crown rot pathogens

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#### Summary

Strawberry powdery mildew (SPM) and Colletotrichum crown rot (CCR), caused by Podosphaera aphanis and Colletotrichum *gloeosporioides*, respectively, are recurring diseases in Florida that can greatly impact yield if not controlled. Nurseries and strawberry growers typically rely on multiple fungicide applications to deliver disease-free transplants and fruit. Increased applications are known to select for resistant populations of pathogens, and result in lack of disease control. Thus, the objectives of this proposal were (i) to screen C. gloeosporioides isolates for sensitivity to the most common fungicides used for CCR management; and (ii) to develop a method to screen and monitor fungicide sensitivity of strawberry powdery mildew.

#### Methods

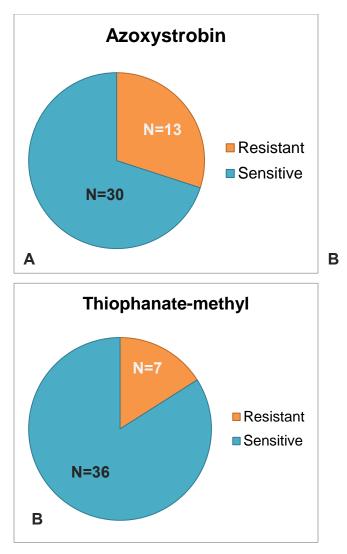
**Objective 1.** During the 2015-16 season, sixty-three *C. gloeosporioides* isolates collected from strawberry crowns between 1995 and 2015 were evaluated for sensitivity to thiophanate-methyl (Topsin) and azoxystrobin (Abound). Preliminary results showed an increased frequency of resistance to azoxystrobin of *C. gloeosporioides* isolates collected after 2007 and reduced in vitro sensitivity to thiophanate-methyl of all the isolates tested. During the 2016-17 season, new isolates were recovered from symptomatic wilting strawberry plants received by the Plant Diagnostic Clinic at the GCREC, in Balm. The crowns of the plants were cut open and a piece of the symptomatic crown was plated on general isolation media. Isolates identified as *C. gloeosporioides* were

transferred onto Potato Dextrose Agar (PDA) and tested for their sensitivity to thiophanate-methyl (Topsin) and azoxystrobin (Abound).

Objective 2. During the 2015- 2016 and 2016- 2017 strawberry seasons, transplants from different cultivar/nursery combinations were transplanted under a tunnel in a research field at the GCREC. SPM symptomatic leaves were collected and DNA was extracted from powdery mildew colonies. Molecular primers were developed to detect whether P. aphanis isolates are resistant to three different groups of fungicides: Demethylation inhibitors (DMIs, i.e., Rally, Procure, Mettle), Quinone-outside inhibitors (QoIs, i.e., Cabrio, Abound), and Succinatedehydrogenase inhibitors (SdhIs, i.e. Fontelis, Kenja, Merivon). A new platform, amplicon sequencing (AmpSeq), is being used to detect the presence and the frequency of point mutations that result in fungicide resistance.

#### Results

**Objective 1.** Forty-three *C. gloeosporioides* isolates recovered from strawberry crowns in 2016-17 were evaluated for sensitivity to azoxystrobin and thiophanate-methyl. Thirty percent (n=13) of the isolates were resistant to azoxystrobin (Abound) (Fig. 1A), whereas 16% (n=7) were resistant to thiophanate-methyl (Topsin) (Fig. 1B). All isolates resistant to thiophanate-methyl were also resistant to azoxystrobin. Thus, fungicides from different chemical groups need to be identified to control CCR in Florida strawberry fields.



**Figure 1.** Sensitivity profile of *Colletotrichum* gloeosporioides isolates from strawberries to **A.** azoxystrobin (Abound) and **B.** thiophanate-methyl (Topsin) at 3 or  $100 \mu$ g/ml.

**Objective 2.** <u>2015 -2016 strawberry season</u>. Data from the AmpSeq analysis confirmed the presence of *P. aphanis* isolates resistant to the DMI and QoI fungicides. Strawberry transplants harboring resistant

isolates originated from the eight nurseries tested. Further investigation is needed to elucidate whether resistant isolates were present in plants before transplanting or if cross-infection between plots occurred within our experimental area. <u>2016 – 2017 strawberry season</u>. *P. aphanis* isolates collected during the 2016 – 2017 season will be analyzed with the AmpSeq platform in August 2017 at the Cornell Institute of Biotechnology (Ithaca, NY). With the results from the AmpSeq and the history of fungicide use in nursery operations, we will attempt to link resistance profiles with each nursery.

### Disclaimer

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