



Orchid fleck dichorhavirus: a new *Brevipalpus*-transmitted virus fresh from Florida



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Chlorotic ringspot symptoms on Giant Lilyturf (*Liriope* spp., cv. 'Gigantea')



Chlorotic leaves on Cast Iron Plant (*Aspidistra elatior*)



Chlorotic spots on *A. elatior* - Early infection?



What was causing these symptoms?

- Tested negative for begomovirus, potyvirus, tospovirus as well as for *Impatiens necrotic spot virus*, *Tobacco mosaic virus* and *Tomato spotted wilt virus*
- Sent to the Florida Department of Agriculture and Consumer Services (FDACS) → sent to USDA



Symptoms on variegated Ophiopogonae

Confirmation: *Orchid fleck dichorhavirus* (OFV)

- Identified using OFV specific conventional reverse transcription polymerase chain assay (RT-PCR) assay + Sanger sequencing + quantitative RT-PCR (RT-qPCR)
- Presence of Orchid fleck dichorhavirus confirmed using OFV generic R2-Dicho-GF and R2-Dicho-GR primers (Roy et al. 2020)
- Sanger sequencing of RT-PCR amplicons shared 98% nucleotide identity with orchid strains of OFV: OFV-Orc1 and OFV-Orc2 (GenBank Accession numbers: AB244418 and LC222630) (Kondo et al. 2006, 2017).

What is OFV?

- Type member for *Dichorhavirus*, family Rhabdoviridae
- Other dichorhaviruses: *Citrus chlorotic spot virus*, *Citrus leprosis virus N*, *Clerodendrum chlorotic spot virus* and *Coffee ringspot virus*
- OFV infects 50+ plant species, including Orchidaceae, Asparagaceae (Nolinoidaea), and Rutaceae (*Citrus*)
- Flat mites from the genus *Brevipalpus* Donnadieu (Trombidiformes: Tenuipalpidae) only known vectors of dichorhaviruses

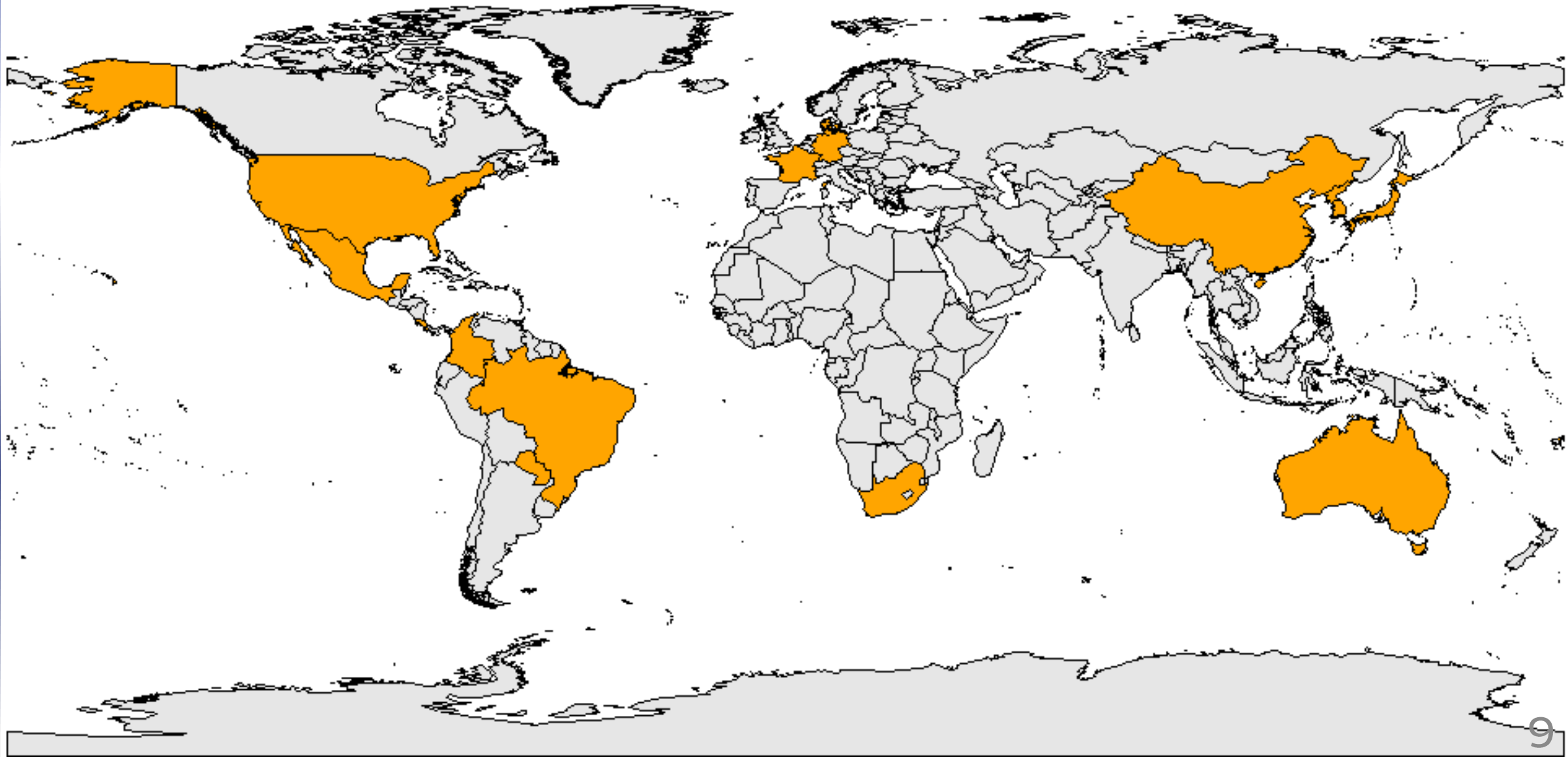


Symptoms of OFV on *Oncidium*

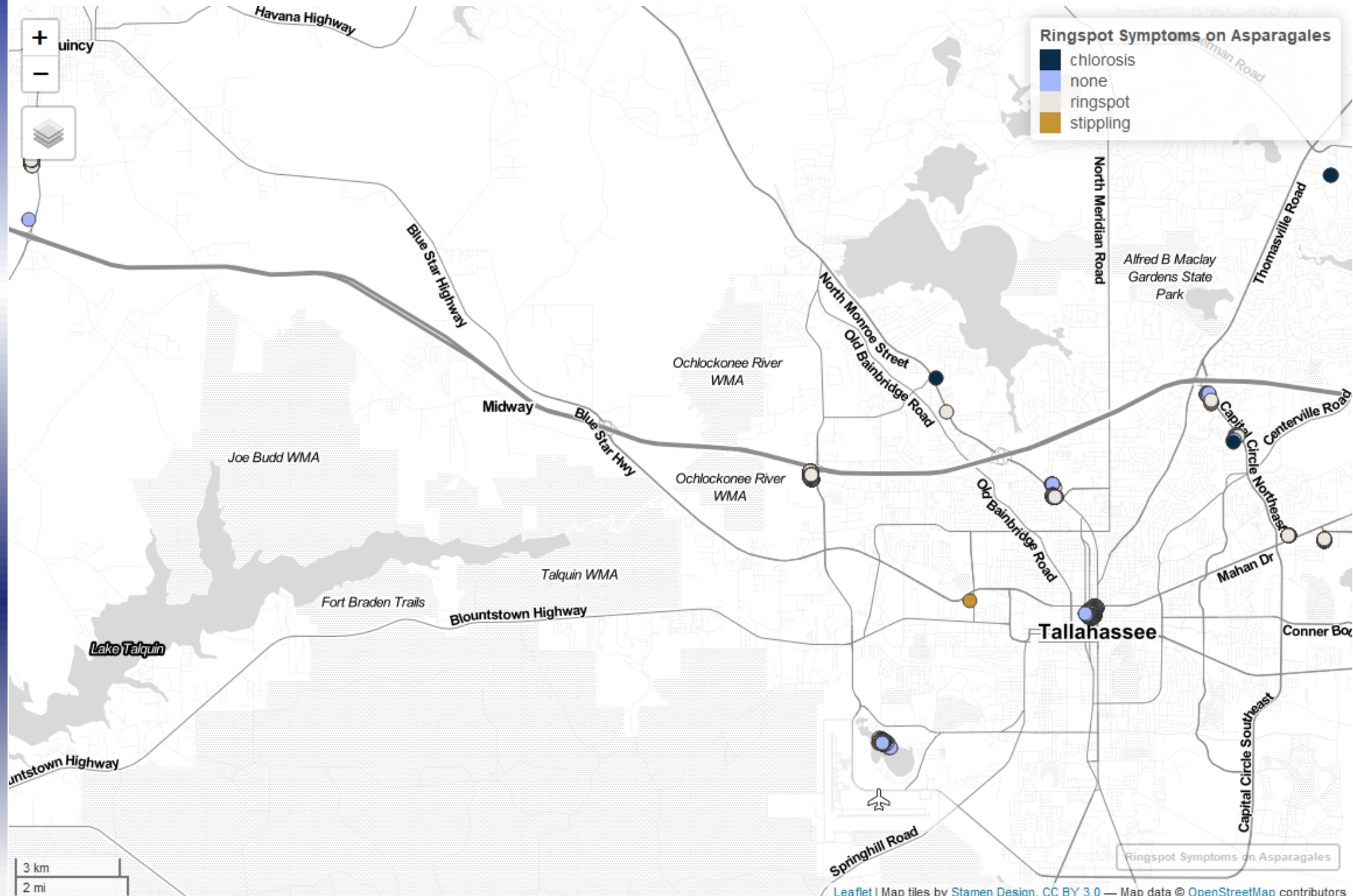
Worldwide Distribution of OFV

- First described infecting *Cymbidium* orchids in Japan (Doi et al. 1977)
- OFV and OFV-like rhabdoviruses have been reported worldwide
- Asia, Africa, North America, South America, Europe and Oceania
- Prevalence of OFV and its mite vector is thought to be associated with the movement of infected orchids (Dietzgen et al. 2018)

World Map of OFV Distribution

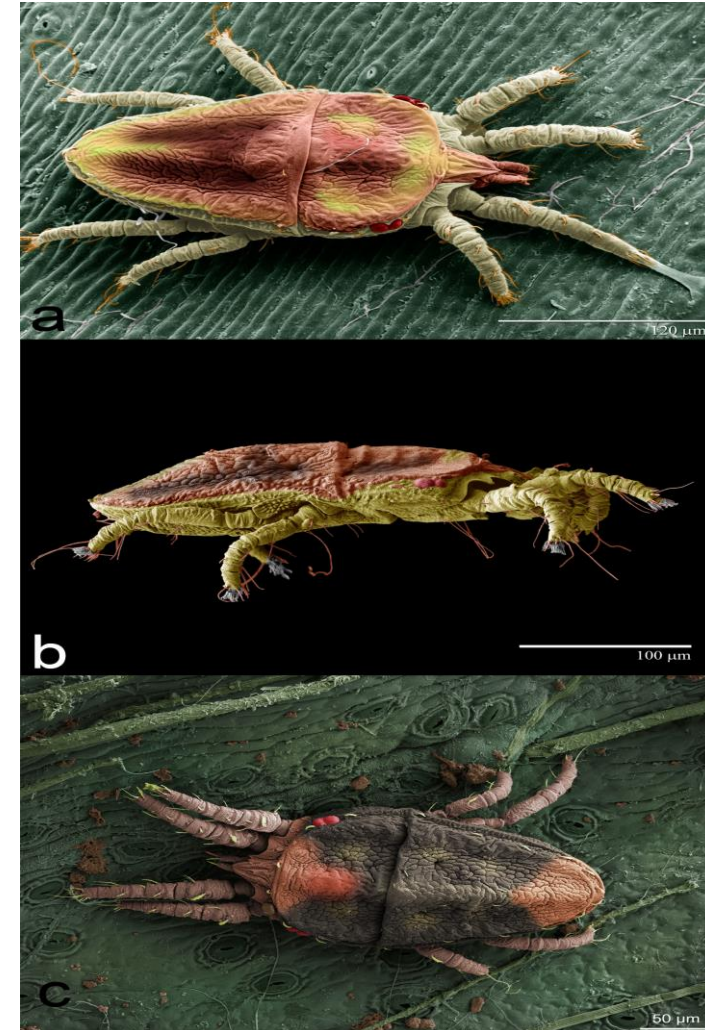


Map of OFV distribution in Tallahassee



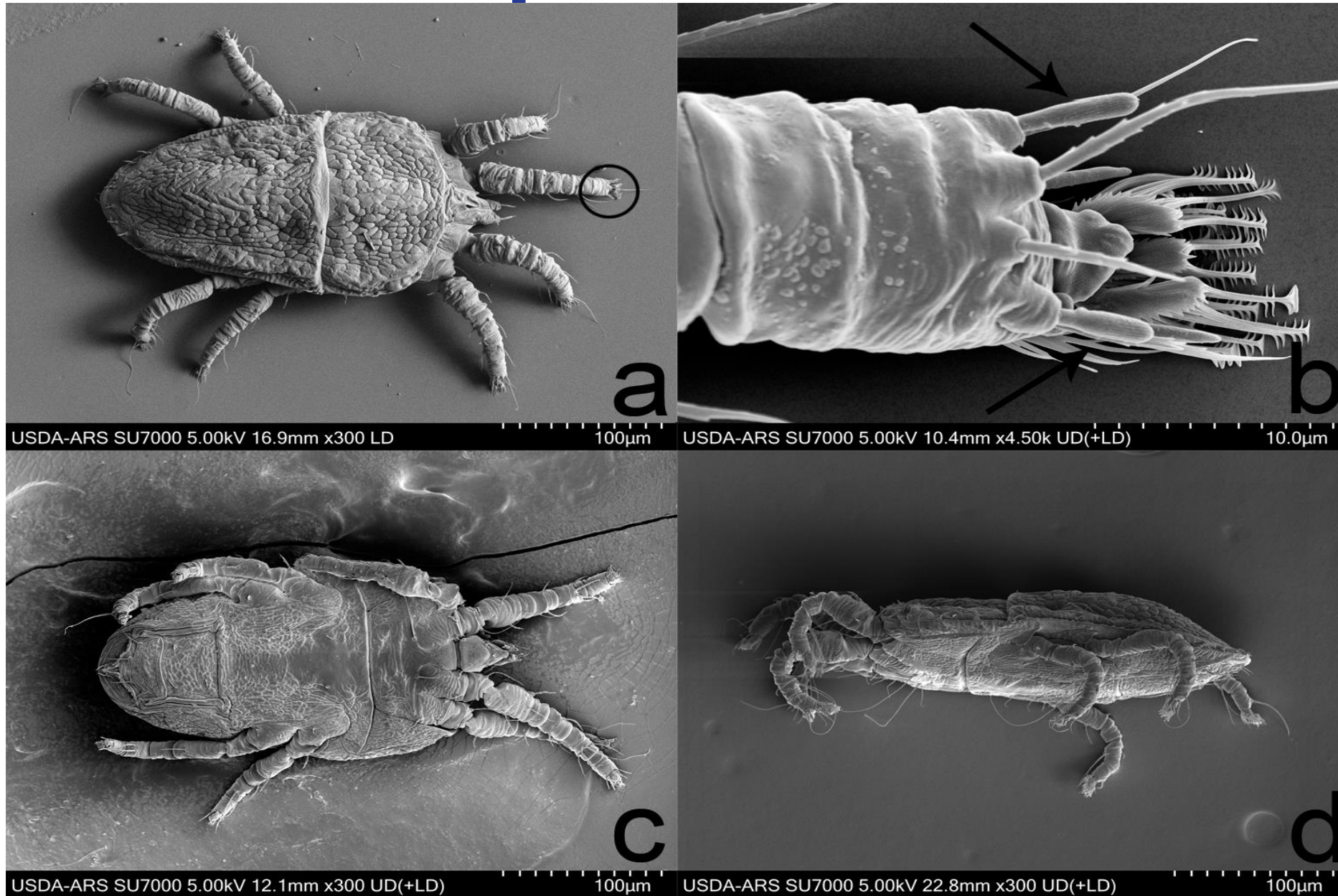
Mites Recovered During Our Survey

- Unidentified eriophyid mite
- Three *Brevipalpus* mite species were recovered from OFV-infected plants:
 - *Brevipalpus californicus* sensu lato
 - *B. obovatus* Donnadieu
 - *B. confusus* Banks
- One of these species is presumably responsible for OFV transmission
- **Cryo-SEM to help determine *Brevipalpus* species identity**



Various *Brevipalpus* spp. live in Florida

The Main Suspect: *B. californicus* s. l.



a. Dorsal view b. Paired solenidia c. Ventral d. Lateral

- *Brevipalpus californicus* (Banks) group are the only known to transmit OFV in a persistent propagative manner - *Brevipalpus* mite species complex contains cryptic species → develop molecular methods?



OFV is an issue for Florida agriculture

- The dichorhavirus that infects citrus in Hawaii, Mexico, Colombia, and South Africa are identical to the OFV in gene order, content, and the genome sequence.
- ICTV: Officially, OFV consists of two orchid strains (OFV-Orc1 and OFV-Orc2) and two citrus strains (OFV-Cit1 and OFV-Cit2)
- Orchid strains of OFV infects citrus (Roy et al. 2020), but none of the citrus strains have been reported from any orchid species
- OFV in Florida represents a concern for horticulturists who grow orchids, *Liriope*, *Ophiopogon*, or other susceptible Asparagaceae species which are commonly used in landscaping



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Orchid fleck virus (OFV)

INTRODUCTION

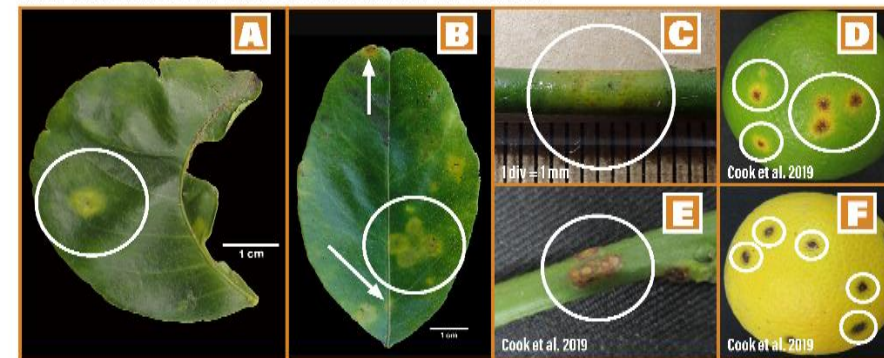
Orchid fleck virus (OFV) causing yellow lesions on leaves and stems was found infecting rough lemon and mandarin orange trees in February 2020 on Hawai'i Island. The virus causes yellow or black leaf lesions on a wide variety of hosts. In 1977, OFV was first described infecting *Cymbidium* sp. leaves in Japan (Doi et al. 1977). Since then, OFV has been reported worldwide infecting more than 70 plant species (Peng et al. 2013).

FIRST REPORT

Leaf and twig samples collected at the University of Hawai'i, College of Tropical Agriculture and Human Resources (UH-CTAHR), Waikeke Research Station were examined by UH-CTAHR researchers who found them to be infected with OFV. This finding was confirmed by scientists at the United States Department of Agriculture (USDA) in Beltsville, MD, who further characterized the virus as OFV orchid strain 2 (OFV-orc2). This confirmation represents new records for Hawai'i and the United States. All known infected trees have been destroyed, and the area will be monitored for any signs of recurrence.

SYMPTOMS

The most notable symptom of OFV are yellow or black bullseye lesions on leaves (images A & B), twigs (images C & E), and fruits (images D & F). These lesions are mostly circular (image A) and can fuse if close together (image B). As lesions age they may change in color from yellow to brown or black. Each lesion represents a feeding site by *Brevipalpus* mites (flat mites) that carry and transmit the virus. Symptom development and display differs between hosts and varieties (Cook et al. 2019; Peng et al. 2013).



OFV lesion symptoms on citrus. **A**, Rough lemon leaf with solitary lesion (circle); **B**, Rough lemon leaf with solitary (arrows) and fused (circles) lesions; **C**, Rough lemon stem with yellow lesion; **D**, Unripe Delta Valencia orange with lesions (circles); **E**, Delta Valencia orange stem with brown lesion (circle); **F**, Ripe Midnight Valencia orange with lesions (circles).

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Page 1 of 2

Plant Pest Control Branch
HAWAII DEPARTMENT OF AGRICULTURE

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