First Report of Phyllocoptes fructiphilus Keifer (Eriophyidae), the Vector of the Rose Rosette Virus, in Florida, USA

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First report of *Phyllocoptes fructiphilus* Keifer (Eriophyidae), the vector of the rose rosette virus, in Florida, USA

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Phyllocoptes fructiphilus Keifer (Acari: Eriophyidae) is a microscopic eriophyid mite. Eriophyid mites are very host specific (Oldfield 1996; Skoracka et al. 2009) and *P. fructiphilus* feeds only on plants in the genus *Rosa* (Amrine 1996). *Phyllocoptes fructiphilus* is associated with the rose rosette emaravirus (rose rosette virus) and acts as the only known vector of rose rosette virus. Infection is associated commonly with the following symptoms: witches' broom, rosetting, deformed flowers, increased thorn density, elongated shoots, reddened leaves and stems, and increased die-back that ultimately kills the rose host (Amrine 1996) (Fig. 1A, B). This disease is known as rose rosette disease and is the most serious illness of roses, affecting the US commercial rose industry which is worth millions of dollars. Rose rosette disease and *P. fructiphilus* have invaded the southeastern US as they followed the range expansion of the non-native *Rosa multiflora* (Thunb.) (Rosaceae) towards the east coast (Amrine 2002; Otero-Colina et al. 2018).

In 2013, a nursery in Quincy, Gadsden County, Florida, USA, detected witches' brooms and other rose rosette disease symptoms on 15 knockout roses that had been imported from out of state. Eight symptomatic plants were tested and found to be positive for rose rosette disease, but *P. fructiphilus* was not detected on the roses at that time (Babu et al. 2014). In 2018, we began a series of surveys along the borders of northern Florida and southern Georgia to determine if this mite was present and acting as a vector for the disease.

Survey efforts initially focused on counties around Leon County, Florida. Rose tissue samples were taken from the periphery of various roses in the landscape; sampling was focused on the flowering tips of roses and included a mixture of flowers, fruits, buds, and short lengths of rose cane. The average sample contained 26.8 ± 1.5 g of undried plant tissue. Samples were trimmed with bypass pruners, dried plant tissue with 70% ethanol between cuts and stored in quart sized plastic bags (Ziploc®, S.C. Johnson & Son, Racine, Wisconsin, USA). Rose cultivars, species, and coordinates were recorded to map out sites that had predatory mites, eriophyid mites, or possible rose rosette disease.

Samples were processed using a washing method derived from Monfreda et al. (2007); cut roses were soaked in a 500 mL beaker with a solution of 1:1 bleach:water with a few drops of concentrated liquid dish washing detergent. The solution was stirred vigorously with a glass rod to dislodge any mites, then poured over a stack of sieves with decreasing screen sizes: 180 µm, 53 µm, and 25 µm. The beaker and rose pieces were further rinsed with tap water over the sieve stack to dislodge any remaining mites. The 53 µm and 25 µm sieves were processed separately; the 53 µm sieve retained larger mites while the 25 µm sieve retained smaller mites, including *P. fructiphilus*. The sieves were then backwashed from the underside of their screen with a water-filled wash bottle, starting from the highest point of a sieve and working to the bottom to flush any trapped debris and mites into a 50 mL centrifuge tube for storage and future observation. Samples were observed under a dissecting microscope. Mites found among the plant debris were siphoned off with a glass pipette and subsequently stored in micro-centrifuge containers with 95% ethanol as a preservative. Some specimens were mounted directly into Hoyer's slide mounting media (Hempstead Halide, Inc., Galveston, Texas, USA), dried at 90 °C, then a ring of nail polish was painted over the edges of the coverslip to seal the slide.

On 14 Feb 2019, we found a total of 42 eriophyid mites from 6 samples obtained from Pink Double Knock Out® roses while surveying roses in the landscape in Tallahassee, Leon County, Florida, USA (Fig. 2A). The mites were sent to the Florida Department of Agriculture and Consumer Services, Division of Plant Industry and were identified as *P. fructiphilus* based, among other characters, on the distinctive pattern of ridges on the prodorsal shield (Bauchan et al. 2019) (Fig. 1C, D). Whereas 2 other eriophyid mites *Eriophyes eremus* Druciarek & Lewandowski and *Phyllocoptes adalius* Keifer (both Acari: Eriophyoidea) are found in roses in the central and eastern US, neither of them were found in the samples analyzed. The roses did not show signs or symptoms of rose rosette disease.

On 16 Jul 2019, we conducted an additional survey of 33 sites with Pink Double Knock Out® roses near the initial site of discovery, including the rose sites where *P. fructiphilus* was detected originally (Fig. 2B). Each sample contained more than 50 eriophyid mites, with some samples containing over 300 mites. We compared the samples collected during Feb and Jul in the same locations with a paired *t*-test and found a significant increase in the *P. fructiphilus* population between the 2 sampling dates (see Fig. 1C; $P = 0.001$; $\alpha = 0.05$; df = 4). Mites that were slide mounted were confirmed subsequently as *P. fructiphilus*.

This is the first record for *P. fructiphilus* in Florida. None of the mite-infested roses showed symptoms of rose rosette disease and none tested positive for rose rosette virus based on detection tools devel-

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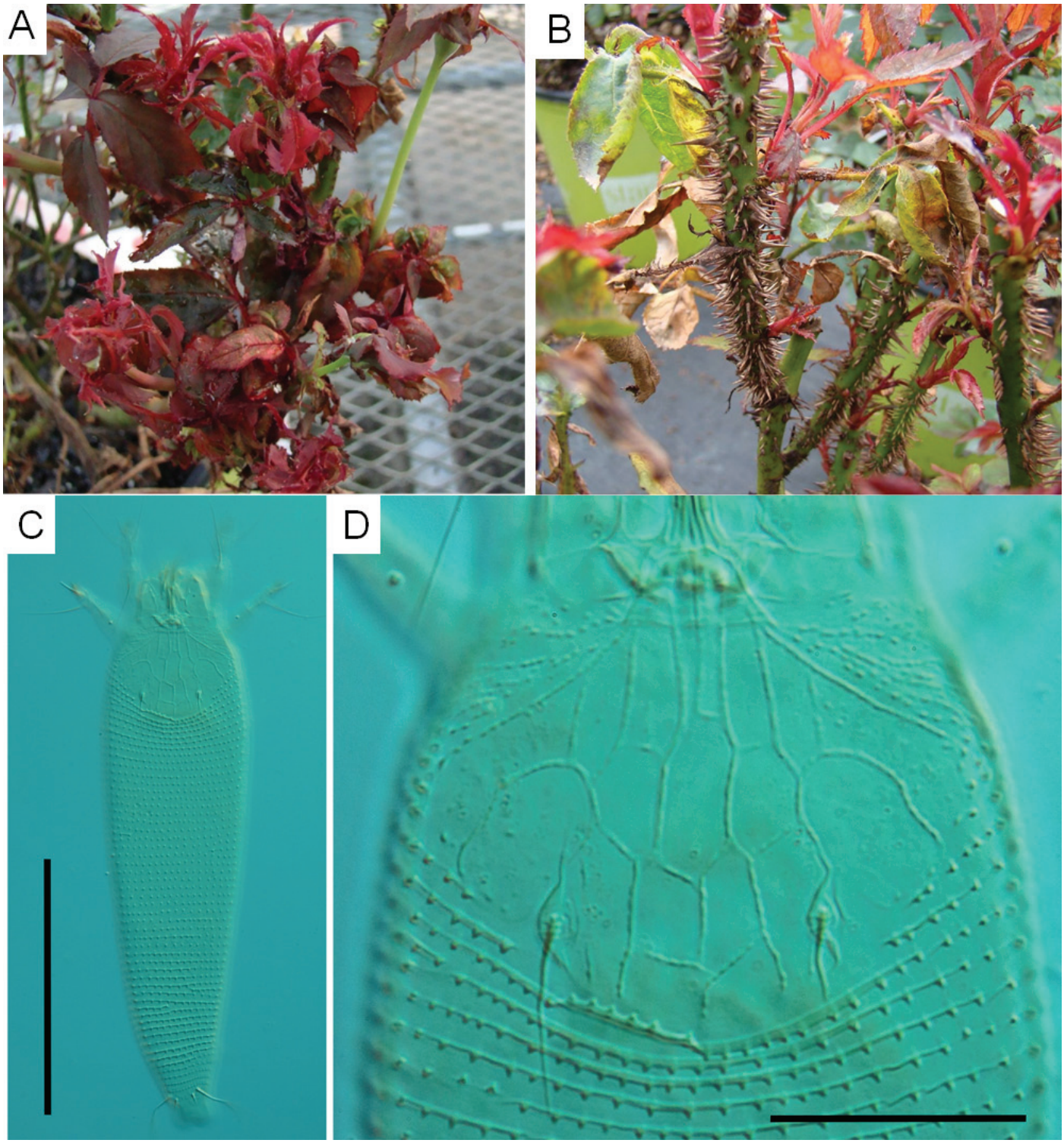

Fig. 1. (A) Symptoms of rose rosette disease: witches' broom, and (B) excessive thorn proliferation; (C) *Phyllocoptes fructiphilus* Keifer (female) from Leon County, Florida, USA: body (scale bar = 100 µm); (D) enlargement of *P. fructiphilus* prodorsal shield to show detail (scale bar = 20 µm).

oped to date. However, the presence of *P. fructiphilus*, along with past detections of rose rosette virus in Florida warrants increased monitoring for the mite and virus in Florida. There is a critical need to develop methods to manage *P. fructiphilus* and rose rosette virus, or homeowners, commercial landscapers, and the US rose industry stands to lose millions of dollars and established plantings in the coming yr.

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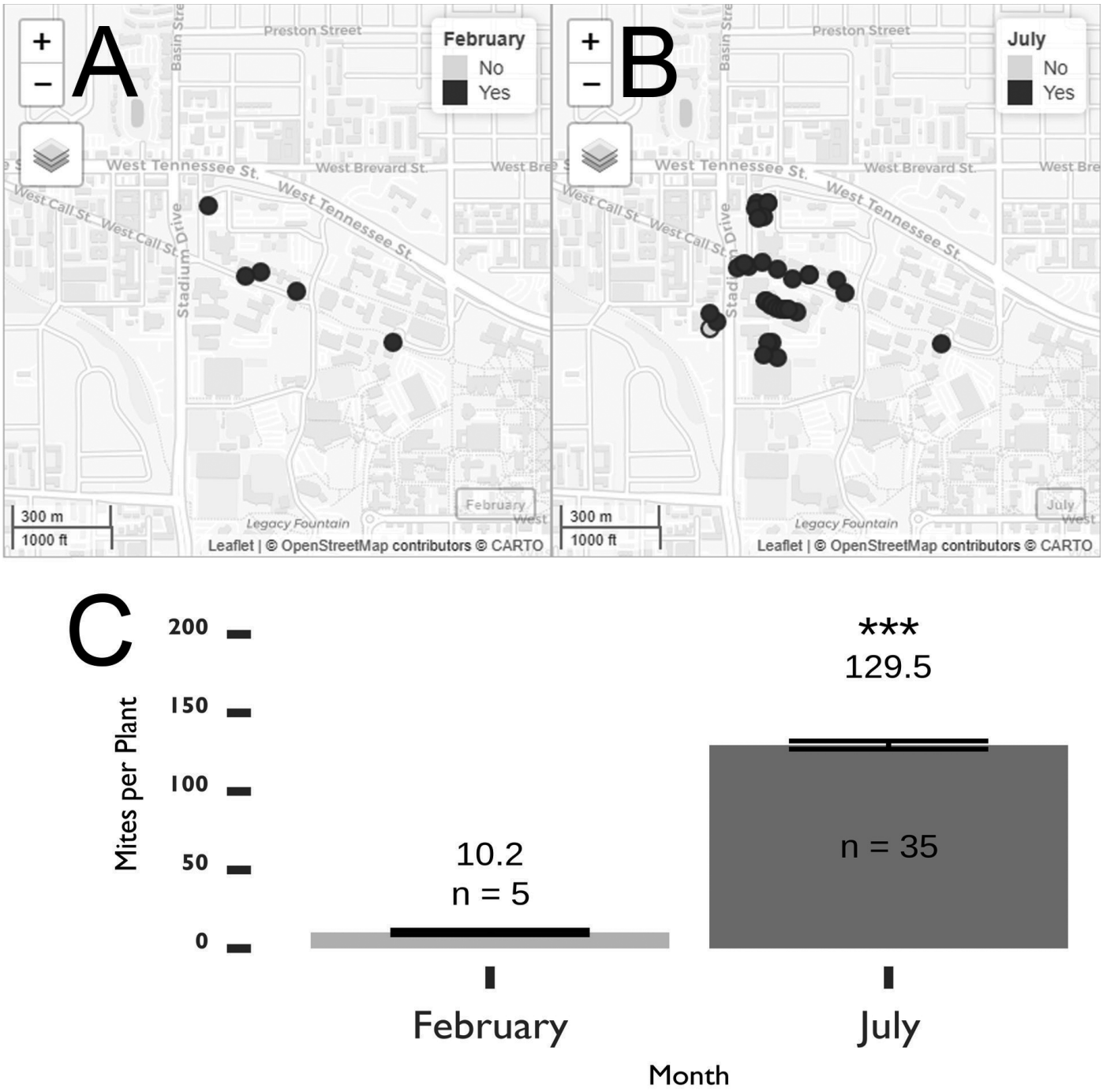

Fig. 2. Presence of *Phyllocoptes fructiphilus* in Leon County, Florida, USA, in (A) Feb 2019 and (B) Jul 2019. Orange dots indicate sites sampled that had *P. fructiphilus*. Gray dots indicate surveyed areas where no *P. fructiphilus* were found. (C) Average number of *P. fructiphilus* per rose sample. Samples were taken from sites in Leon County, Florida, on 14 Feb and 16 Jul 2019. Asterisks represent significant differences as calculated by pairwise *t*-tests of the 5 sites tested for *P. fructiphilus* during both mo. *P*-value < 0.001.

Summary

The invasive mite *Phyllocoptes fructiphilus* Keifer (Acari: Eriophyidae) feeds on plants in the genus *Rosa*. *Phyllocoptes fructiphilus* is associated with the rose rosette emaravirus (rose rosette virus) and acts as the only known vector of rose rosette virus, the causal agent of rose rosette disease (Emaravirus). The mite *P. fructiphilus* is reported for the first time in the state of Florida, USA. No roses showed signs or symptoms of viral infection, and current molecular methods were unable to detect the virus. *Phyllocoptes fructiphilus* represents a potential threat to the Florida rose industry if rose rosette disease becomes established.

Key Words: rose rosette disease; rose rosette virus

Sumario

El ácaro invasivo *Phyllocoptes fructiphilus* Keifer (Acari: Eriophyidae) se alimenta sobre plantas del género *Rosa. Phyllocoptes fructiphilus* se asocia con rose rosette emaravirus (virus del arrosetamiento de la rosa), es reconocido principalmente como vector de la virus del arrosetamiento de la rosa, el agente causal de la enfermedad del arrosetamiento de la rosa (Emaraviridae). El ácaro *P. fructiphilus* se reporta por primera vez para el estado de la Florida, USA. Ninguna rosa mostró señales o síntomas de una infección viral, y ningun virus fue detectado con el uso de métodos moleculares de hoy en día. *Phyllocoptes fructiphilus* representa una amenaza potencial para la industria de la rosa en la Florida si Emaraviridae se llega a establecer.

Palabras Clave: virus del arrosetamiento de la rosa; enfermedad del arrosetamiento de la rosa; emaravirus

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