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Field survey of Asian citrus psyllid (Hemiptera: Liviidae) infestations associated with six cultivars of *Poncirus trifoliata* (Rutaceae)

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The Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Liviidae), is an important pest because it transmits a bacterium ‘*Candidatus Liberibacter asiaticus*’ putatively responsible for a serious citrus disease known as Asiatic huanglongbing or citrus greening disease (Bové 2006; Gottwald 2010). Classic recommendations to growers confronted with the disease are to plant disease-free nursery stock, routinely identify and remove infected trees to reduce inoculum loads, and aggressively manage populations of the psyllid (Hall et al. 2013). Insecticidal control is the key tactic used to manage the psyllid, but host plant resistance may hold some promise in the search for alternative tactics. Whereas no resistance to the psyllid has been observed within the *Citrus* genus (Rutaceae), relatively strong levels of antixenotic resistance to the psyllid have been reported in *Poncirus trifoliata* (L.) Raf. (Rutaceae), a species in the same Rutaceae subfamily as *Citrus*. Aubert (1987) noted reduced infestations of the psyllid on *P. trifoliata*. Westbrook et al. (2011) conducted a field survey of 87 genotypes in the plant family Rutaceae and concluded *P. trifoliata* was one of the most resistant to psyllid colonization. Recent laboratory and greenhouse investigations confirmed that *P. trifoliata* cultivars usually are colonized less by the psyllid than are *Citrus* cultivars (Richardson & Hall 2013; Hall et al. 2015).

The purpose of the research presented here was to assess infestations of the psyllid associated with 5-yr-old *P. trifoliata* trees under field conditions. Infestation densities of eggs and nymphs on flush shoots

were monitored over the summer of 2016 on 6 pure *P. trifoliata* cultivars, 4 citrange cultivars, and 2 conventional *Citrus* cultivars—a sweet orange (*Citrus sinensis* [L.] Osbeck) cultivar (Hamlin) and a sweet orange hybrid (Temple) (Table 1). Citranges are hybrids between *P. trifoliata* and sweet orange. Six to 8 trees of each of the 12 cultivars were available for monitoring in a large planting of many experimental citranges (0.8 ha area, 8 rows with about 100 trees per row, 1.5 m spacing between trees, 1 cultivar per row). The trees were subjected to regular irrigation, fertilization, and weed control but without any insecticides or horticultural oils. On each sample date, 2 flush shoots (each with at least some leaves appropriate for oviposition) were randomly collected from each tree, transported to a laboratory, and examined under a microscope to count numbers of eggs and nymphs of *D. citri*. Additionally on each sample date, each tree was examined to estimate the percentage of branches with flush shoots appropriate for oviposition. Data on log-transformed numbers of psyllids per flush shoot and on arcsine-transformed percentages of flush shoots with 5th instar nymphs were subjected to analyses of variance (PROC GLM; SAS Institute 2010), mean comparisons among cultivars were investigated with the Ryan-Einot-Gabriel-Welsch multiple range test, and results were reported with untransformed means.

Poncirus trifoliata is a deciduous genotype, and trees in the field are thus usually completely void of foliage throughout the winter,

Table 1. Mean number of immature Asian citrus psyllids per flush shoot in a 5-yr-old field planting of *Citrus*, citrange, and *P. trifoliata* trees sampled during Mar to Sep 2016.

Genotype group	Cultivar	Mean number per flush shoot ^a		Mean percentage of infested samples with fifth instar nymphs ^a
		Eggs	Nymphs	
<i>Citrus</i>	Hamlin	16.0ab	21.6a	37.2ab
<i>Citrus</i>	Temple	10.6b	22.5a	47.7ab
Citrange	C-35	25.0a	20.4a	18.5ab
Citrange	Carrizo	18.6ab	16.1a	23.3ab
Citrange	Norton	21.4ab	20.2a	34.0ab
Citrange	Uvalde	28.7ab	24.4a	35.3ab
<i>Poncirus</i>	Argentina	0.8c	1.5b	13.3b
<i>Poncirus</i>	Flying Dragon	2.0c	1.1b	7.7b
<i>Poncirus</i>	Large Flower	0.8c	0.8b	57.1a
<i>Poncirus</i>	Pomeroiy	0.8c	1.6b	27.3ab
<i>Poncirus</i>	Rich 16-6	0.9c	0.4b	20.0ab
<i>Poncirus</i>	Rubidoux	1.7c	1.0b	9.1b

^aMeans in the same column followed by the same letter are not significantly different ($P > 0.05$), Ryan-Einot-Gabriel-Welsch multiple range test.

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whereas citranges may be semi-deciduous. During early Mar of this survey, ample quantities of flush shoots were present in trees of the *Citrus* and citrange cultivars, and psyllid eggs and nymphs were present on this flush. However, none of the *P. trifoliata* trees at this time had yet broken winter dormancy and thus all of them were barren of foliage. The *P. trifoliata* cultivars began to flush toward the end of Mar. Thereafter during the summer, at least some flush was consistently available on the *P. trifoliata* cultivars, and flushing patterns were similar in *Citrus*, citranges, and *P. trifoliata* (Fig. 1a). Among 9 sample dates during late Mar to Sep, relatively large infestation densities of eggs and nymphs were observed on the *Citrus* and citrange cultivars whereas significantly fewer were consistently observed on the pure *P. trifoliata* cultivars (Table 1; Fig. 1b). Over all sample dates, means \pm SE of 13.3 ± 1.9 , 23.7 ± 2.4 , and 1.1 ± 0.2 eggs per flush shoot were observed on the *Citrus*, citrange, and *P. trifoliata* cultivars, respectively. Although fewer eggs were deposited on *P. trifoliata*, at least some of these hatched: means of 22.0 ± 2.3 , 20.8 ± 1.9 , and 1.0 ± 0.2 nymphs per flush shoot were observed on the *Citrus*, citrange, and *P. trifoliata* cultivars, respectively. Few nymphs were observed on the pure *P. trifoliata* shoots but at least some developed to the 5th instar (Table 1), and general observations indicated these older nymphs were healthy enough that they would have successfully molted to the adult stage.

Reduced colonization by Asian citrus psyllid on *P. trifoliata* was largely a result of reduced rates of oviposition. Because *P. trifoliata* readily hybridizes with *Citrus* species, if the specific traits responsible for reduced oviposition can be identified, then it might be possible to transfer these traits to conventional *Citrus* cultivars. However, we found no reduced oviposition on the 4 citrange cultivars tested in this study. The resistance in *P. trifoliata* to psyllid oviposition may be either a recessive or a multi-genic trait, or there may be genetically controlled traits in *Citrus* that promote oviposition.

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Summary

The Asian citrus psyllid (Hemiptera: Liviidae) transmits a bacterium responsible for huanglongbing, a serious citrus disease. Insecticidal control of the psyllid is a key tactic used to manage the disease, but host plant resistance may hold some promise and clues in the search for alternative tactics. Results of a field survey revealed that relatively large infestation densities of the psyllid developed on conventional *Citrus* (Rutaceae) and citrange (Rutaceae) cultivars but not on any of 6 *Poncirus trifoliata* (L.) Raf. (Rutaceae) cultivars. *Poncirus trifoliata* is a species closely related to *Citrus*, and citranges are hybrids of sweet orange and *P. trifoliata*. Reduced colonization by the psyllid on *P. trifoliata* was largely a result of reduced rates of oviposition. *Poncirus trifoliata* resistance to oviposition was not observed in 4 citrange cultivars.

Key Words: citrus greening; huanglongbing; *Diaphorina citri*; Liberibacter

Sumario

El psílido asiático de los cítricos (Hemiptera: Liviidae) transmite la bacteria responsable del huanglongbing, una enfermedad seria de los cítricos. El control químico del psílido es una táctica clave utilizada para manejar la enfermedad, pero la resistencia de la planta hospedera puede ser prometedora y dar una pista en la búsqueda de tácticas alternativas. Los resultados de un sondeo del campo revelaron densidades de infestación relativamente grandes del psílido desarrollándose en cultivares convencionales de *Citrus* (Rutaceae) y citrange (Rutaceae), pero no sobre ninguno de los 6 cultivares de *Poncirus trifoliata* (L.) Raf. (Rutaceae). *Poncirus trifoliata* es una especie estrechamente relacionada con *Citrus* y las citranges son híbridos de naranja dulce y *P. trifoliata*. La reducción de la colonización por el psílido sobre *P. trifoliata* fue en gran parte resultado de una tasa de oviposición reducida. No se observó resistencia a la oviposición en *Poncirus trifoliata* en los 4 cultivares de citrange.

Palabras Clave: enverdecimiento de los cítricos; huanglongbing; *Diaphorina citri*; Liberibacter

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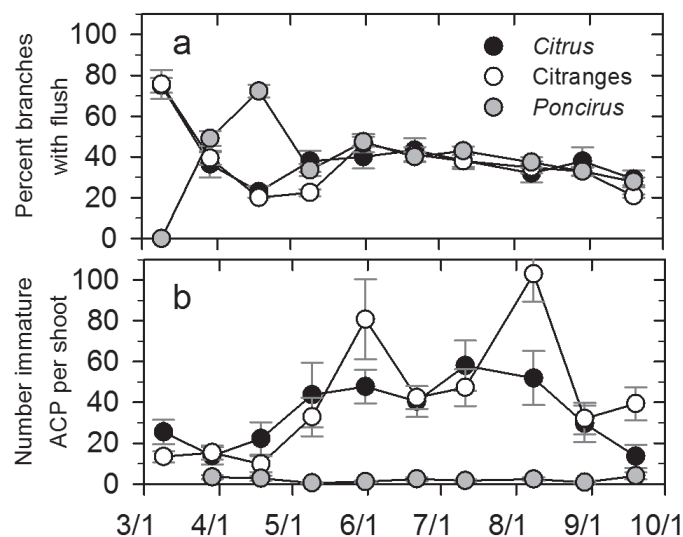


Fig. 1. Comparisons among cultivars of *Citrus*, citranges, and pure *Poncirus trifoliata* with respect to infestations of Asian citrus psyllid (ACP) in 5-yr-old trees at a grove in east-central Florida during 2016. a) Percentage of branches with flush suitable for oviposition by Asian citrus psyllid. b) Average infestation densities of immature Asian citrus psyllids (counts of eggs and nymphs combined) per flush shoot. Error bars are standard errors of the mean.