Abundance of Citrus Leafminer Larvae on *Citrus* and *Citrus*-related Germplasm

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Abstract. The citrus leafminer, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae), is a key pest in most citrus-growing regions worldwide. Adult citrus leafminers oviposit primarily on young elongating flush of *Citrus* as well as other Rutaceae and some ornamental plants. Larvae feed on the epidermal cell layer of developing leaves and injury to leaves provides a pathway for infection by the bacterium *Xanthomonas citri* subsp. *citri* (Hasse), the causal agent of Asiatic citrus canker. In this study, we quantified abundance of citrus leafminer larvae on progeny of 87 seed parent genotypes of *Citrus* and *Citrus* relatives (family Rutaceae) in the field in East-central Florida to identify those that have low abundance of leafminers. Progeny from the 87 parent genotypes varied in abundance of the leafminer. Progeny of 15 parent genotypes had a high mean abundance of more than six leafminers per flush shoot. All but one of these genotypes were in the *Citrus* genus. Progeny of 16 parent genotypes had zero, or nearly zero, leafminers, but none were from the *Citrus* genus. However, many of these 16 genotypes were from genera closely related to true citrus (subtribe Citrinae) and are sexually compatible with *Citrus*. Progeny of two parent genotypes in the subfamily Toddalioidae and *Glycosmis pentaphylla* (Retz.) Corr. also had a low abundance of leafminer. *Glycosmis pentaphylla* also is a poor host for the Asian citrus psyllid, *Diaphorina citri* Kuwayama, and has biochemical resistance to the citrus weevil, *Diaprepes abbreviatus* (L.), so this genotype as well as others identified as poor hosts for the leafminer may prove useful in breeding programs aimed at reducing the abundance of multiple insect pests on citrus.

The citrus leafminer, *Phyllocnistis citrella*, is native to Southeast Asia but has become a key pest in most citrus-growing regions worldwide (Heppner, 1993). The citrus leafminer was first discovered in major citrus-producing states in the United States (i.e., California, Florida, and Texas) in the 1990s and early 2000s and has rapidly spread throughout these states and

elsewhere (Heppner, 1993; Legaspi et al., 1999). Adult citrus leafminers oviposit primarily on young elongating flush of all varieties of *Citrus* as well as other Rutaceae and some ornamental plants (Heppner, 1993; Jacas et al., 1997; Pandey and Pandey, 1964). Larvae feed on the epidermal cell layer of developing leaves, producing a serpentine mine (Belasque et al., 2005). Leafminer-damaged leaves may become curled and twisted and heavy infestations of the insect can stunt the growth of plants and reduce yield (Peña et al., 2000). The citrus leafminer also causes wounds to leaves that can provide a pathway for infection by the bacterium *Xanthomonas citri* subsp. *citri*, which causes Asiatic citrus canker and reduces the quality and quantity of fruit (Chagas et al., 2001; Christiano et al., 2007; Hall et al., 2010).

Current management of the citrus leafminer is largely by insecticides and classical biological control in the United States (Hoy and Nguyen, 1997; Hoy et al., 2007; Pomerinke and Stansly, 1998; Powell et al., 2007; Sétamou et al., 2010). However, control of larvae with

foliar insecticides is often ineffective because larvae are protected from residues within their mines and also may develop resistance to a broad range of insecticides (Villanueva-Jiménez and Hoy, 1998). Biweekly applications of insecticides may be needed to protect emerging, highly susceptible leaf flush, but these insecticides reduce the populations of natural enemies of the leafminer (Smith and Peña, 2002). Classical biological control agents are sometimes effective at reducing populations of citrus leafminer; however, their effectiveness is tempered by environmental conditions (Yoder and Hoy, 1998). The population of natural enemies also lags behind the pest population and, therefore, may provide minimal control early in the growing season (Hoy et al., 2007). Development of effective alternatives to insecticides and classical biological control for management of the citrus leafminer is of critical importance. Host plant resistance ultimately may provide the most effective, economical, environmentally safe, and sustainable method of control, especially if the plant also is resistant to other important pests of citrus.

Traits that confer resistance to insects have been documented among members of the orange subfamily Aurantioideae (Bowman et al., 2001; Luthria et al., 1989; Yang and Tang, 1988). Very little is known about antixenosis or antibiosis of *Citrus* and relatives to the leafminer. Only two relatives, *Berbera koenigii* L. and *Glycosmis pentaphylla*, are known to be completely resistant to the leafminer (Fletcher, 1920; Jacas et al., 1997), but genotypes of *Citrus* and relatives may differ in susceptibility (Batra and Sandhu, 1983; Heppner, 1993; Jacas et al., 1997; Wilson, 1991), and in some cases, the mechanism conferring avoidance or resistance has been identified (Batra and Sandhu, 1983; Batra et al., 1984; Bernet et al., 2005; Jacas et al., 1997; Padmanaban, 1994; Singh et al., 1988). In this study we quantified abundance of citrus leafminer larvae on progeny of seed parent genotypes of *Citrus* and *Citrus* relatives (family Rutaceae) in the field in East-central Florida to identify those that have low abundance of leafminers. Ultimately, *Citrus* and relatives that have low abundance of the leafminer may have mechanisms that lower the population and economic impact of this pest.

Materials and Methods

We obtained seeds from 124 accessions of *Citrus* and relatives in the family Rutaceae (subfamily Aurantioideae) from the USDA-ARS National Clonal Germplasm Repository for Citrus and Dates located at the University of California at Riverside (UCR). The seeds were collected from the Citrus Variety Collection of UCR (see <<http://www.citrusvariety.ucr.edu>>), which was created nearly 100 years ago and contains more than 1100 accessions [each with a unique Citrus Research Center (CRC) number]. Among the seeds from the 124 accessions from the Citrus Variety Collection was the Core Collection of Citrus hybrids, which represents ≈85% of the genetic

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Table 1. Mean abundance of larval citrus leafminer on *Citrus* and *Citrus* relatives in Ft. Pierce, FL.^z

Botanical name of seed parent ^y (CRC ^x)	Common name of seed parent ^y	No.	Mean rank ^w	Mean abundance (0–3) ^v
<i>C. maxima</i> (Burm.) Merr. (3805)	Reinking	28	468.3 ^a	2.9
<i>C. neo-aurantium</i> (<i>C. obovoidea</i> + <i>C. unshiu</i> graft chimera) (3816) ^u	Kinkoji unshiu graft chimera	28	456.9 ^{ab}	2.8
<i>C. aurantium</i> L. (2717) ^u	Olivelands sour orange	26	454.5 ^{abc}	2.8
<i>C. paradisi</i> Macf. (3781) ^u	Tahitian pummelo × star ruby grapefruit	28	446.7 ^{abcd}	2.8
<i>C. excelsa</i> Wester (2317) ^u	Limon real (papeda)	29	439.9 ^{abcd}	2.7
<i>C. hassaku</i> , hort ex Tan. (3942) ^u	Hassaku pummelo hybrid	30	439.6 ^{abcd}	2.7
<i>C. maxima</i> (Burm.) Merr. (2248) ^u	Kao panne pummelo	30	433.1 ^{abcde}	2.7
<i>C. limon</i> (L.) Burm. f. (3885) ^u	Lemon variety from Iran	28	427.4 ^{abcdef}	2.6
<i>C. taiwanica</i> Tan. & Shimada (2588)	Nansho daidai sour orange	29	418.0 ^{abcdefg}	2.6
<i>C. nobilis</i> Lour. (3845) ^u	King tangor	26	416.7 ^{abcdefg}	2.6
<i>C. maxima</i> (Burm.) Merr. (4026) ^u	Pomelit pummelo hybrid	29	410.5 ^{abcdefgh}	2.5
× <i>Citroncirus</i> sp. (<i>C. paradisi</i> ‘Duncan’ × <i>P. trifoliata</i>) (3771)	Swingle citrumelo	32	408.2 ^{abcdefgh}	2.5
<i>C. aurantium</i> L. (3929)	Gou tou cheng	31	407.6 ^{abcdefgh}	2.5
<i>C. medica</i> L. (661)	Indian citron hybrid	26	406.8 ^{abcdefgh}	2.5
<i>C. maxima</i> (Burm.) Merr. (2242) ^u	Kao pan pummelo	28	403.7 ^{abcdefgh}	2.4
<i>C. limon</i> (L.) Burm. f. (3593) ^u	Interdonato lemon	30	403.2 ^{bcdefgh}	2.4
× <i>Citrofortunella</i> sp. (3172)	Tavares limequat	29	400.7 ^{bcdefghi}	2.4
<i>C. maxima</i> (Burm.) Merr. (3959) ^u	Egami buntan pummelo	32	396.0 ^{bcdefghij}	2.4
<i>C. reticulata</i> Blanco (3558) ^u	Fremont mandarin	26	395.9 ^{bcdefghijk}	2.4
<i>C. hassaku</i> hort ex Tan. (3907) ^u	Hassaku pummelo hybrid	26	394.1 ^{bcdefghijk}	2.4
<i>C. macrophylla</i> Wester (3842) ^u	Alemow	30	392.5 ^{bcdefghijk}	2.4
<i>C. volkameriana</i> / <i>C. limonia</i> Osbeck (3050)	Volkamer lemon hybrid	26	391.8 ^{bcdefghijkl}	2.4
<i>C. davaoensis</i> (Wester) Tan. (2427)	Davao lemon (papeda)	27	391.2 ^{bcdefghijkl}	2.3
<i>C. limon</i> L. Burm. f. (3005) ^u	Frost nucellar eureka lemon	26	388.5 ^{cdefghijklm}	2.3
<i>C. limon</i> (L.) Burm. f. (3892) ^u	Mesero lemon	28	385.8 ^{defghijklm}	2.3
<i>C. reticulata</i> Blanco (3022) ^u	Frua mandarin	28	385.6 ^{defghijklm}	2.3
<i>C. neo-aurantium</i> Tan. (3611) ^u	Konejime sour orange hybrid	26	384.7 ^{defghijklmn}	2.3
<i>C. reticulata</i> Blanco (2590) ^u	Tien chieh mandarin	27	382.4 ^{defghijklmno}	2.4
<i>C. limonia</i> (L.) Osbeck (712)	Santa barbara red lime	31	382.1 ^{defghijklmno}	2.3
<i>C. jambhiri</i> Lush. (400)	Florida rough lemon	30	379.8 ^{defghijklmno}	2.3
<i>C. aurantium</i> L. (3930)	Zhuluan sour orange hybrid	23	378.6 ^{defghijklmnop}	2.3
<i>C. reticulata</i> hybrid (Clementine × Orlando) (3851) ^u	Lee mandarin	23	369.4 ^{efghijklmnopq}	2.3
<i>C. reticulata</i> hybrid (Clementine × Orlando) (3850) ^u	Robinson mandarin	31	366.7 ^{fghijklmnopq}	2.2
<i>C. reticulata</i> Blanco (3812) ^u	Unnamed mandarin	28	366.4 ^{fghijklmnopq}	2.1
<i>C. benikoji</i> hort. ex Tan. (3149)	Unnamed	26	364.6 ^{fghijklmnopq}	2.2
<i>C. reticulata</i> Blanco (3260)	Soh niamtra mandarin	31	363.9 ^{fghijklmnopq}	2.1
<i>C. reticulata</i> Blanco (3958) ^u	Koster mandarin	29	362.5 ^{fghijklmnopq}	2.1
<i>C. reticulata</i> Blanco (3018)	Dweet tangor	30	362.3 ^{fghijklmnopq}	2.2
<i>C. reticulata</i> Blanco (300)	Parson’s special mandarin	28	361.7 ^{fghijklmnopq}	2.1
<i>C. limonia</i> (L.) Osbeck (3919) ^u	Lamas rangpur lime	29	357.6 ^{ghijklmnopq}	2.1
<i>C. sunki</i> hort. ex Tan. (3143)	Sunki mandarin	30	357.6 ^{ghijklmnopq}	2.1
<i>C. reticulata</i> Blanco (3752) ^u	Som keowan mandarin	28	357.2 ^{ghijklmnopq}	2.1
<i>C. limettoides</i> Tan. (1482) ^u	Palestine sweet lime	24	355.5 ^{ghijklmnopqr}	2.2
<i>C. medica</i> L. (3546) ^u	South coast field station citron	29	345.6 ^{hijklmnopqr}	2.0
<i>C. intermedia</i> hort. ex Tan. (3474)	Yama-mikan sour orange	27	345.1 ^{hijklmnopqs}	2.1
<i>C. aurantiifolia</i> (Christm.) Swing. (2450) ^u	India lime	29	336.4 ^{ijklmnopqrst}	2.0
<i>C. limon</i> (L.) Burm. f. (3176) ^u	Frost nucellar Lisbon lemon	27	334.3 ^{jklmnopqrst}	2.0
<i>C. longispina</i> Wester (2320) ^u	Talamisan	28	332.5 ^{jklmnopqrst}	2.0
× <i>Citroncirus</i> sp. (3552)	S-281 citrangelo	29	331.7 ^{klmnopqrst}	1.9
<i>C. reticulata</i> Blanco (4003)	Sun chu sha mandarin	28	329.8 ^{klmnopqrst}	1.9
<i>C. medica</i> L. (3523)	Diamante citron	31	328.1 ^{lmnopqrst}	1.9
<i>C. hybrid</i> (53-1-16 ‘Clem’ × ‘Hamlin’) × <i>Chinotto</i> F1 (3715)	Sour orange hybrid ex-india	25	323.1 ^{lmnopqrst}	2.0
<i>C. sinensis</i> (L.) Osbeck (3858)	Pineapple sweet orange	29	322.6 ^{mnpqrst}	1.9
<i>C. reticulata</i> (3326)	Scarlet emperor mandarin	27	317.6 ^{nopqrst}	1.9
<i>C. aurantium</i> L. (628)	Standard sour orange	28	316.2 ^{opqrst}	1.9
<i>C. aurantiifolia</i> (Christm.) Swing. (3822) ^u	Mexican lime type	29	313.1 ^{pqrst}	1.8
<i>C. amblycarpa</i> Och. (2485) ^u	Nasnaran mandarin	29	309.7 ^{qrst}	1.8
<i>C. maxima</i> (Burm.) Merr. (3945) ^u	Mato buntan pummelo	22	299.7 ^{qrstu}	1.7
× <i>Citroncirus</i> sp. (301)	Rusk citrange trifoliolate hybrid	30	288.4 ^{rstu}	1.6
<i>C. lycopersiformis</i> hort. ex Tan. (3564)	Monkey orange	28	288.4 ^{rstu}	1.6
<i>C. latipes</i> (Swing.) Tan. (3052)	Khasi papeda	27	279.5 ^{stu}	1.6
<i>Severinia buxifolia</i> (Poiret) Tan. (1497)	Chinese box orange	25	272.9 ^{tu}	1.5
<i>C. reticulata</i> Blanco (3363) ^u	Belady mandarin	27	265.4 ^{tu}	1.5
<i>C. webberi</i> Wester (1455) ^u	Kalpi papeda	30	263.2 ^{tuv}	1.4
<i>C. leiocarpa</i> hort. ex Tan. (3147)	Koji mandarin	26	253.9 ^{tuvw}	1.5
<i>C. aurantium</i> L. (3289) ^u	Sour orange var. salicifolia	10	246.1 ^{tuvwx}	1.3
<i>Poncirus trifoliata</i> L. (4007)	“Little-leaf” trifoliolate	31	232.9 ^{uvwxy}	1.2
× <i>Citroncirus</i> sp. (<i>Cleopatra mandarin</i> × <i>trifoliata</i>) (3957)	X639 trifoliolate hybrid	31	200.6 ^{wxyz}	1.0
<i>C. halimii</i> B.C. Stone (3780) ^u	Unnamed	26	192.6 ^{wxyz}	1.0
<i>Afraegle paniculata</i> (Schum.) Engl.	Nigerian powder flask fruit	31	190.2 ^{wxyz}	0.9
<i>Microcitrus inodora</i> (F.M. Bail) Swing. (3785)	Large leaf australian wild lime	17	172.7 ^{xyzA}	0.8
<i>Microcitrus</i> hybrid (<i>M. australis</i> × <i>M. australasica</i>) (1485)	Sydney hybrid	31	136.9 ^{yzAB}	0.5

(Continued on next page)

Table 1. (Continued) Mean abundance of larval citrus leafminer on *Citrus* and *Citrus* relatives in Ft. Pierce, FL.^z

Botanical name of seed parent ^y (CRC ^x)	Common name of seed parent ^y	No.	Mean rank ^w	Mean abundance (0–3) ^v
<i>Poncirus trifoliata</i> L. (3549) ^a	Simmons trifoliolate	28	136.7 ^{yzAB}	0.5
× <i>Microcitronella</i> sp. (<i>M. australasica</i> × Calamondin) (1466)	Faustrimedin	26	133.1 ^{zAB}	0.5
<i>Balsamocitrus dawei</i> Stapf (3514)	Uganda powder-flask	38	132.0 ^{zAB}	0.4
<i>Microcitrus australis</i> (Planch.) Swing. (3673)	Australian round lime	26	120.3 ^{AB}	0.4
<i>Eremocitrus glauca</i> (Lindley) Swing. Hybrid (4105)	Australian desert lime hybrid	30	118.5 ^{AB}	0.3
<i>Aegle marmelos</i> (L.) Corr.	Indian bael fruit	27	95.0 ^B	0.1
<i>Glycosmis pentaphylla</i> (Retz.) Corr. (3285)	Orangeberry/gin berry	32	94.7 ^B	0.1
<i>Bergera koenigii</i> L. (3165)	Curry tree	31	92.6 ^B	0.1
<i>Microcitrus australasica</i> (F.J. Muell.) Swing. (1484)	Australian finger lime ‘Sanguinea’	22	86.8 ^B	0.0
<i>Casimiroa edulis</i> Llave et Lex (-)	White sapote	32	85.7 ^B	0.0
<i>Murraya paniculata</i> L. (3171)	var. ovatifoliolata	31	83.8 ^B	0.0
<i>Murraya paniculata</i> (L.) Jack (1637)	Orange Jessamine	29	82.9 ^B	0.0
<i>Clausena excavata</i> Burm.f. (3166)	Pink wampee	32	81.1 ^B	0.0
<i>Zanthoxylum ailanthoides</i> L. (-)	Japanese prickly-ash	31	79.7 ^B	0.0
<i>Clausena harmandiana</i> (Pierre) Guillaumin (4034)	Unnamed	27	78.4 ^B	0.0

^aMembers of the Rutaceae vary greatly in their incidence of nucellar embryony, so some of the plants tested were essentially genetically identical to the seed parent, whereas others were sexual hybrids.

^bBotanical and common names specified by Citrus Variety Collection, Riverside, CA (<http://www.citrusvariety.ucr.edu>).

^cAccession number assigned by Citrus Research Center and maintained by the Citrus Variety Collection.

^dMean rank was calculated using a nonparametric Friedman’s test and ranks with different letters (or capitalization) are significantly different.

^eAbundance of larvae was quantified on a 0 to 3 ordinal scale: 0 = no larvae; 1 = one to two larvae; 2 = three to six larvae; and three to six or more larvae.

^fFrom the Core Collection of Citrus hybrids, which represents ≈85% of the genetic diversity at the Citrus Variety Collection.

diversity of the UCR collection (Barkley, 2003). We also obtained seed from the subfamily Aurantioideae, *Afraegle paniculata* (Schum.) Engl. and *Aegle marmelos* (L.) Corr., and the subfamily Toddalioidae, *Casimiroa edulis* (Llave et Lex) and *Zanthoxylum ailanthoides* (L.), from the Fruit and Spice Park (Miami/Dade County, FL) and the University of Georgia. Members of the Rutaceae vary greatly in their incidence of nucellar embryony (reviewed in Frost and Soost, 1968); therefore, some of the plants we tested were genetically identical to the female parent, whereas others were sexual hybrids. Phylogenetic relationships of the seed parent genotypes (hereafter “parent genotypes”) we used are described in Barkley (2003) and Bayer et al. (2009).

We planted seeds of each parent genotype in individual plastic cells (3.8 × 21 cm) (SC-10 super cell Cone-tainers; Stuewe and Sons, Corvallis, OR) containing sterile potting mix. Seedlings of 87 parent genotypes were successfully propagated in a greenhouse at the USDA-ARS U.S. Horticultural Research Laboratory in Fort Pierce, FL. We transplanted seedlings to 3.7-L containers 4 to 7 months after sowing and grew them in a greenhouse with a mean diurnal temperature cycle of 35 °C maximum and 23 °C minimum in the summer and 32 and 20 °C in the winter. We watered plants daily and fertilized them weekly.

After growing plants for 6 to 9 months in the greenhouse, we planted one seedling of each parent genotype in each of eight randomized complete blocks on a research farm owned by the USDA-ARS (Fort Pierce, FL) during June and July 2009. An additional plant derived from *Balsamocitrus dawei* Staph (CRC 3514) was planted in each of two blocks. Trees were planted in three rows with 0.6-m spacing between trees in a row and 3.5 m between rows. We regularly irrigated and fertilized trees using a program similar to that used for new commercial plantings of citrus. No pesticides were applied during the study. We surveyed the

trees four times, at approximately monthly intervals from June to Sept. 2010, to measure the abundance of citrus leafminer larvae.

We estimated abundance of leafminers only on trees with a minimum of one flush shoot because larvae are dependent on flush to feed and develop. A tree was omitted during a survey time if no flush was present. Flush was defined as any shoot with developing leaves, which included newly breaking buds to shoots with expanded but tender young leaves (Hall and Albrigo, 2007). Morphology of plants and flush differed, which necessitated careful examination of each plant to locate flush. We sampled progeny from all parent genotypes on each date, but obtained the maximum number of samples (32 or 40 across all dates) for only five genotypes (Table 1). The mean number of samples per parent genotype was 28 as a result of the lack of synchronous flushing by plants on our sampling dates. For plants with multiple flush shoots, the first shoot found to contain leafminers was used to determine abundance for that replicate. Abundance of larvae was estimated visually and quantified on a 0 to 3 ordinal scale: 0 = no larvae; 1 = one to two larvae; 2 = three to six larvae; and 3 = more than six larvae. Whereas integer counts likely would identify smaller effects of genotype on abundance of leafminer, categorical counts were adequate to meet our objective and necessary to sample all replicates in a reasonable time period. Our counts do not directly consider the severity of the mines. Most of the mines appeared well developed, and the number of days mining as well as the number of larvae per leaf is correlated with damage (Schaffer et al., 1996), so our counts are likely correlated with severity of the mines.

Statistical analyses. We tested whether the abundance of citrus leafminer differed among seedlings of the 87 parent genotypes using a non-parametric repeated-measures analysis:

the F-approximation of the Friedman test (Ipe, 1987) and the associated rank sum multiple comparison test (PROC GLM; SAS Institute, 2008).

Results and Discussion

Abundance of citrus leafminer larvae varied among progeny of the 87 parent genotypes of citrus and citrus relatives ($F = 22.9$, $df = 86$, $P < 0.001$; Table 1). Progeny of 15 parent genotypes averaged nearly 3 on our ordinal scale, meaning they had more than six larvae per flush shoot (Table 1). All of these genotypes, except for ×*Citroncirus* sp. (CRC 3771), were in the genus *Citrus*. Progeny of many parent genotypes in the genus *Citrus* had a moderate to low abundance of leafminer. However, wounds caused by the leafminer are susceptible to infection by the canker bacterium for a relatively long period of time and require only a small concentration of inoculum (Christiano et al., 2007), so even a low abundance of leafminer may increase the rate of infection of a tree.

Progeny of 16 parent genotypes had zero, or nearly zero, leafminers, and none of these were in the genus *Citrus* (Table 1). However, this group includes genotypes from other genera of true citrus (subtribe Citrineae) that are sexually compatible with *Citrus* (Swingle, 1943; Swingle and Reece, 1967): *Microcitrus* hybrid (CRC 1485); *Poncirus trifoliata* ‘Simmons trifoliolate’ (CRC 3549); ×*Microcitronella* sp. (CRC 1466); *M. australis* (3673); *Eremocitrus glauca* (CRC 4105); and *M. australasica* (CRC 1484). *Poncirus trifoliata* is the only species in this group previously identified as having some resistance to the leafminer, but we used a different cultivar than the previous study (Bernet et al., 2005). There are four major groups of *Poncirus* (Fang et al., 1997) and our limited data on this genus suggest that abundance of leafminer larvae may vary

among large-flowered and small-flowered genotypes (Table 1). *P. trifoliata* is graft-compatible with *Citrus*, is used as rootstock in many citrus-growing regions (Krueger and Navarro, 2007; Ziegler and Wolfe, 1981), and is an important parent in intergeneric hybrids with *Citrus* (Krueger and Navarro, 2007). Therefore, *P. trifoliata* may be useful in breeding programs as a potential source of genes that confer resistance to insects.

Progeny from additional parent genotypes had a low or zero abundance of leafminer, including those in the subfamily Toddalioidae, *Casimiroa edulis* and *Z. ailanthoides*, and two genotypes previously identified as resistant to the leafminer, *B. koenigii* and *G. pentaphylla* (Fletcher, 1920; Jacas et al., 1997). *Glycosmis pentaphylla* is a remote citroid fruit (Swingle, 1943; Swingle and Reece, 1967) and is sexually incompatible with species in the genus *Citrus*. However, genes identified in *G. pentaphylla* that prove to confer resistance to insects could be transferred to cultivated varieties of citrus using transgenic or intragenic methods (Rommens et al., 2007). *Glycosmis pentaphylla* also is an unfavorable host to the Asian citrus psyllid and has biochemical resistance against the citrus weevil, *Diaprepes abbreviatus* (L.) (Shapiro et al., 1997, 2000). We did not test the influence that colonization by other insect species such as the Asian citrus psyllid had on abundance of citrus leafminer. The Asian citrus psyllid concurrently infested many of these trees and probably colonizes new flush earlier than the citrus leafminer. However, competition between the insect species was not apparent because the patterns of abundance of the two species were similar across progeny of the parent genotypes (Westbrook et al., 2011). For example, in addition to *G. pentaphylla*, the following genotypes had low abundances of both insect species: *Aegle marmelos*, *C. edulis*, *Clausena hammandiana*, *E. glauca*, *M. australasica*, a *Microcitrus* hybrid (CRC 1485), *P. trifoliata* 'Simmons trifoliolate', and *Z. ailanthoides* (Westbrook et al., 2011). Therefore, these genotypes may be useful in breeding programs aimed at reducing populations of multiple insect pests.

In conclusion, the majority of the progeny we evaluated supported a moderate to high abundance of citrus leafminer, confirming the broad range of hosts for this insect pest within the Aurantioideae (Heppner, 1993; Jacas et al., 1997; Pandey and Pandey, 1964). However, some true citrus and citrus relatives had virtually no larval leafminers, which indicates that they may lack cues that attract the leafminer, may not be preferred in a choice situation, or may possess mechanisms that confer resistance to the leafminer. Resistance may result from morphological or chemical defenses that deter leafminers or reduce their fitness. In our study we used young trees in a polyculture and attractiveness or resistance in some plant species to insects may vary as a result of the age of the plant or attributes of neighboring plants (Smith, 2005). Identifying whether resistance is expressed by these genotypes throughout the lifespan of the tree and in

monoculture and identifying genes that confer resistance are the next steps toward developing citrus varieties that limit leafminer populations and, indirectly, the spread of Asiatic citrus canker.

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