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Bionomics of Asian Citrus Psyllid (Hemiptera: Liviidae) Associated with Orange Jasmine Hedges in Southeast Central Florida, with Special Reference to Biological Control by *Tamarixia radiata*

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ABSTRACT The Asian citrus psyllid, *Diaphorina citri* Kuwayama, is an important pest in Florida because it transmits bacteria responsible for citrus huanglongbing disease. In addition to infesting citrus, orange jasmine (*Murraya exotica* L.) is one of Asian citrus psyllid's preferred host plants and is widely grown as an ornamental hedge. We report on Asian citrus psyllid bionomics over three years at five urban plantings of orange jasmine and on biological control of Asian citrus psyllid by a parasitoid *Tamarixia radiata* (Waterston). *T. radiata* had been released in Florida shortly after Asian citrus psyllid was first found, and the parasitoid was known to be established at each planting. Additionally, three new *T. radiata* haplotypes were released every 3 wk at three plantings during the first study year (one haplotype per planting, over all releases an average of 17 parasitoids per linear meter of hedge); all three haplotypes were released at a fourth planting beginning midway through the study (over all releases, an average combined total of 202 parasitoids per linear meter of hedge). Asian citrus psyllid populations were present year-round at each planting, often at large levels. Such plantings may pose risk to commercial citrus as Asian citrus psyllid reservoirs. Releases of the new haplotypes did not cause any measurable reduction in Asian citrus psyllid population levels during the study, and ironically percentage parasitism was generally highest at a planting where no releases were made. Higher release rates might have been more effective. The probability is discussed that repetitive pruning of orange jasmine reduced the full potential of *T. radiata* against Asian citrus psyllid in this study.

KEY WORDS greening disease, huanglongbing, *Murraya*, *Diaphorina citri*

The Asian citrus psyllid, *Diaphorina citri* Kuwayama, is an important invasive citrus pest in the United States because it transmits a bacterium "*Candidatus* Liberibacter asiaticus" (CLas) responsible for a devastating disease of citrus known as Asiatic huanglongbing (HLB) (Bové 2006, Hall et al. 2013a). Asian citrus psyllid and HLB were first found in Florida in 1998 and 2005, respectively, and the disease has since put this state's citrus industry in serious jeopardy. Mature citrus trees infected by the HLB bacterium become unthrifty with thinning canopies, the fruit from such trees are smaller and may be unmarketable due to reduced size and a bitter taste, and infected trees may drop fruit before harvest; young citrus trees that become infected generally decline and die faster than older trees (Gottwald 2010, Hall and Gottwald 2011). The citrus acreage in Florida has been significantly reduced due to HLB, and replanting trees and getting them into production without contracting HLB is difficult even under intensive Asian citrus psyllid insecticide programs (Hall et al. 2013b). The seriousness of HLB and roadblocks to finding solutions are complex and multifaceted.

Orange jasmine (*Murraya exotica* L. = *Murraya paniculata* auct. non.) is widely grown as an ornamental hedge plant in Florida and is regarded as one of the psyllid's preferred host plants. After HLB was found in Florida, concern developed that orange jasmine grown in urban areas might contribute to larger area-wide Asian citrus psyllid populations, thereby increasing Asian citrus psyllid populations in nearby commercial citrus. Grower and regulatory concerns about orange jasmine increased when a report was published that orange jasmine can serve as a host of CLas (Damsteegt et al., 2010). Subsequent research indicated that Asian citrus psyllid coming off urban plantings of orange jasmine may only play a minor role in spreading CLas to citrus because the pathogen was rarely detected in either orange jasmine or Asian citrus psyllid collected from orange jasmine (Walters et al. 2012a,b); thus, the major concern about orange jasmine may be its influence on area-wide Asian citrus psyllid population levels.

Shortly after the discovery of the psyllid in 1998, a parasitoid of Asian citrus psyllid, *Tamarixia radiata*

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(Waterston) (Hymenoptera: Eulophidae), was imported from Southeast Asia (Taiwan and South Vietnam) and released in Florida citrus (Hoy et al. 1999, Hoy and Nguyen 2001). The parasitoid established and has since been reported to occur widely in Florida citrus, but its populations in citrus have been somewhat sporadic usually causing relatively low levels of Asian citrus psyllid parasitism (Qureshi et al. 2009), even in orchards not treated with insecticides (Hall et al. 2008). In an effort to boost biological control of Asian citrus psyllid by *T. radiata* in Florida, the Florida Department of Agriculture and Consumer Services (Division of Plant Industry—DPI) and University of Florida imported three different geographic populations of *T. radiata* and, after permits were granted, urgently began releasing these in Florida during 2011 (Hall et al. 2013a). The populations were imported from south China, North Vietnam, and Pakistan—Barr et al. (2009) showed that the populations represented a single species but that they could be distinguished based on mitochondrial COI data and coded them as haplotypes 1, 4, and 5, respectively. USDA-ARS collaborated with DPI by making releases of the three haplotypes at urban plantings of orange jasmine in southeast central Florida. This provided an opportunity to assess the importance of urban plantings of orange jasmine as reservoirs of Asian citrus psyllid. Also, most commercial citrus in Florida was being subjected to intensive insecticide programs aimed at controlling Asian citrus psyllid and thus not suitable for releases of the parasitoids.

Presented here are the results of a study to assess the bionomics of Asian citrus psyllid at five urban plantings of orange jasmine and to evaluate biological control of Asian citrus psyllid by *T. radiata* at these plantings. The study took place over a 34-mo period during which each planting was visited monthly to record information on Asian citrus psyllid population levels. For one year beginning 3 mo after the study was initiated, repetitive releases were made of the Chinese, Vietnamese, and Pakistan populations of *T. radiata*, each population at one orange jasmine planting. After these releases were discontinued, Asian citrus psyllid and *T. radiata* populations were monitored at the three plantings for an additional 19 mo. An intent of these releases was to provide an opportunity for each haplotype to establish and to reduce Asian citrus psyllid population levels. Sometimes thousands of adult parasitoids were released at a time, and of interest was if such releases resulted in an immediate reduction in Asian citrus psyllid population levels, even if only temporarily. After releases were discontinued at the three plantings, releases were initiated at a fourth planting where large numbers of all three haplotypes were released repetitively throughout the remainder of the study. A fifth planting where none of the *T. radiata* haplotypes were released was continually monitored for Asian citrus psyllid and *T. radiata* over the 34-mo study.

Materials and Methods

Study Sites. Five urban plantings of orange jasmine were surveyed monthly for infestations of Asian citrus psyllid from March 2011 through December 2013. Relatively large, well-manicured hedges of jasmine were present at these sites (see Supp Information 1 [online only]); however, each site was unique most notably with respect to parasitoid releases; thus, data on psyllid populations at the five sites were not directly compared.

Site 1: Gardens (26° 50'54.37" N, –80° 05'6.40" W). Located at an entrance into the Palm Beach Gardens Mall parking lot in the town of North Palm Beach in Palm Beach County, this hedge was 37.2 m long and was maintained at a height of about 0.9 m and a width of 2.1 m; the planting was irrigated and fully exposed to the sun. There were similar hedges at each of four other entrances to the mall, and about half way through our study we had to switch to one of the other plantings about 81 meters to the northwest.

Site 2: Citrus (27° 26'39.82" N, –80° 19'42.75" W). This hedge flanked a parking lot at Citrus Professional Building near downtown Fort Pierce in Saint Lucie County; the hedge was regularly irrigated and much of the hedge was fully exposed to the sun. There were other similar hedges in the vicinity. Details of the size of this hedge are given in Table 1.

Site 3: Jupiter (26° 55'55.16" N, –80° 08'14.42" W). Grown as a hedge dividing two parking lots in the town of Jupiter in Martin County, this planting was heavily shaded by trees and not irrigated. The hedge was initially somewhat weak and progressively became weaker, which was attributed to lack of irrigation, poor soil, too much shade and encroachment of paved parking lots along each side of the hedge. Details of the size of this hedge are given in Table 1.

Site 4: Laurel (27° 24'50.50" N, –80° 20'56.65" W). This hedge flanked a parking lot at Laurel Professional Park between Fort Pierce and White City in Saint Lucie County; the planting was irrigated, and most of the hedge was fully exposed to sun primarily in the afternoon. Details of the size of this hedge are given in Table 1.

Site 5: Kings (27° 19'2.49" N, –80° 23'4.41" W). This hedge flanked the entranceway into a gated community known as King's Isle in the town of Saint Lucie West in Saint Lucie County, it was irrigated and most of the hedge was fully exposed to sun. Details of the size of this hedge are given in Table 1.

The plantings were situated from north-to-south in the following order: Citrus, Laurel, Kings, Jupiter, and Gardens, respectively, with about 76 km from Citrus to Gardens. Laurel was located 4.1 km south of Citrus and 11.4 km north of Kings. Kings was 49 km north of the Jupiter site, and Jupiter was 11 km north of Gardens. Based on satellite imagery and ground truthing, we estimated that the closet commercial citrus to each study site was around 8 km from Citrus, 6 km from Laurel, 9 km from Kings, 6 km from Jupiter, and 17 km from Gardens. A map showing these locations is provided in Supp Information 1 (online only).

Releases of *T. radiata* Haplotypes. The three haplotypes of *T. radiata* were supplied by DPI in Gainesville, FL. At approximately 3-wk intervals, newly emerged adult parasitoids were aspirated in groups of 100 into 50-ml polypropylene tubes (3 cm wide, 11.5 cm

Table 1. Releases of new haplotypes of *T. radiata* at four urban plantings of orange jasmine and the size of the hedge at each planting

Parasitoid release/hedge information	Planting			
	Citrus	Laurel	Jupiter	Kings
Haplotype released	Vietnam	China	Pakistan	All three
Total number parasitoids released	26,100	26,900	26,814	151,456
Number of release dates	20	21	22	28
Average number per release	1,300	1,220	1,280	5,350
Average number per linear meter of hedge	17.2	15.3	18.4	201.9
Average number per square meter of hedge	24.6	17.0	23.0	91.8
Average number per cubic meter of hedge	20.5	15.4	25.6	76.5
Maximum number per release	3,000	2,800	2,300	12,300
Maximum number per linear meter of hedge	39.7	35.0	33.1	464.2
Hedge linear length (m)	75.6	79.9	69.5	26.5
Hedge width (m)	0.7	0.9	0.8	2.2
Upper surface area of hedge (m^2)	52.9	71.9	55.6	58.3
Height of hedge (m)	1.2	1.1	0.9	1.2
Hedge volume (m^3)	63.5	79.1	50.0	70.0

long; Thermo Fisher Scientific, Waltham, MA) provisioned with pure honey as described by Hall and Klein (2014) and then shipped overnight to Fort Pierce, where the adults were released the same day the shipment arrived. The tubes and parasitoids were transported in a cooler to release sites where, after removing the lid, each tube was gently wedged into the upper area of a hedge leaving the tube in an upright position—the adults escaped through the tube's opening and dispersed on their own. Sometimes 20–40 or more tubes with adults were deployed along a hedge, and afterward starting with the first tube deployed we would ensure all parasitoids had escaped (if any remained we would tap them out onto the hedge).

No releases were made at Gardens. The plantings at Citrus, Jupiter, and Laurel were designated as initial release sites for the Vietnamese, Pakistan, and Chinese haplotypes of *T. radiata*, respectively. Releases at these plantings were initiated during May 2011 3 mo after the study began and were continued usually at 3-wk intervals until May 2012 (Table 1). Releases of all three haplotypes were initiated at Kings during June 2012, 16 mo after the study began, and were continued usually every 3 wks until near the end of the study in December 2013. All three haplotypes were released at Kings on each date, although the number of each haplotype usually differed depending on how many of each DPI shipped. Table 1 summarizes the combined releases of the three haplotypes at Kings, with the following specifics for each haplotype: for the Vietnamese haplotype, a total of 62,286 parasitoids were released with an average of 1,950 per release (500 minimum, 5,400 maximum); for the Pakistan haplotype, a total of 31,450 adults were released with an average of 1,300 per release (150 minimum, 3,300 maximum); and for the Chinese haplotype a total of 57,720 adults were released with an average of 2,100 per release (500 minimum, 4,300 maximum). Of the grand total of 151,456 parasitoids released at Kings, 41, 21 and 38% were Vietnamese, Pakistan, and Chinese haplotypes, respectively.

Flush Abundance. The abundance of flush suitable for Asian citrus psyllid oviposition at each planting was estimated monthly by counting the number of flush shoots per 697 cm^2 (0.07 m^2) of foliage along the upper horizontal surface of each hedge using a rectangular frame (22.9- by 30.5-cm opening) made of polyvinyl chloride pipe (1.3 cm in diameter). This rectangular frame was placed at each of 10 locations along the upper hedge surface spanning from one end of a hedge to the other. At each sample location, all flush shoots with at least some feather flush leaves appropriate for oviposition were counted within and below the frame's opening (regularly pruned hedges usually have a canopy of dense, exterior foliage with little or no interior leaf production).

Abundance of Eggs and Nymphs. Each time a planting was visited, 25 flush shoots appropriate for colonization by immatures were examined for Asian citrus psyllid eggs and nymphs. The shoots selected for examination spanned from one end of a hedge to the other. Asian citrus psyllid densities per shoot were estimated using a 0–3 rating system (from Westbrook et al. 2010)—for eggs the ratings were 0 (no eggs present), 1 (1–20 eggs), 2 (21–40 eggs), or 3 (>40 eggs); for nymphs the ratings were 0 (no nymphs present), 1 (1–10 nymphs), 2 (11–30 nymphs), or 3 (>30 nymphs). For each sample date, the percentage of shoots infested by eggs and the percentage infested by nymphs were calculated. No information was collected on the age of nymphs.

Parasitism by *T. radiata*. Biological control of Asian citrus psyllid by *T. radiata* was investigated monthly at each planting by excising flush shoots with older (larger) Asian citrus psyllid nymphs (usually 4th and 5th instars), placing these shoots into a cooler and returning them to a lab. Each flush shoot was inspected to remove any spiders, aphids, lady beetles, or other unwanted arthropods. A 148-ml plastic vial (#8940, BioQuip Products, Rancho Dominguez, CA) filled with water was used to hold flush shoots for emergence of Asian citrus psyllid and parasitoid adults. These vials came with plastic snap-on lids, and we used a razor blade to cut two slits in an X pattern through the center of each lid. The base end of the stem of each shoot was given a fresh diagonal cut with a razor blade and then placed through the junction of the lid's slits into the vial of water. For the first half of the study, 10 flush shoots with older nymphs were collected at each site and held individually for emergence of Asian citrus psyllid and parasitoid adults. Each shoot with its vial of water was slipped down into a polycarbonate Magenta vessel (model GA-7, 7.6 by 7.6 by 10.2 cm, Magenta LLC, Chicago, IL) fitted with a polypropylene coupler (Caisson Laboratories, North Logan, UT), and a second Magenta vessel was then inverted and attached to this coupler (the bottom of the second vessel was modified so that it had a screened hole for ventilation). The vessels containing flush shoots were placed on a lab bench under ambient conditions (22.9°C) ~60 cm below bright, cool LED lights (EnduraLED lamps, 940 lumens, Philips North America, Andover, MA), 14 h of daily illumination. The shoots were held for 2 wks and inspected every 2 or 3 d to remove emerged Asian citrus psyllid and parasitoid adults. Total numbers of adult Asian citrus psyllid and adult *T. radiata* that emerged from each shoot were counted, and apparent percentage parasitism of Asian citrus psyllid associated with each shoot was calculated based on these numbers (McAuslane et al. 1993). All Asian citrus psyllid and *T. radiata* adults that emerged were placed into propylene glycol and stored for genetic analyses to be conducted and reported elsewhere.

During July 2012 in order to increase numbers of adult *T. radiata* collected for genetic analyses, the above procedure was changed from collecting 10 shoots to collecting enough shoots to provide ~100 older Asian citrus psyllid nymphs; these shoots were subdivided into three groups with sometimes as many as 10 shoots per group, the number of shoots per group was recorded, and then each group of flush shoots was placed together through a lid into a vial of water. The coupled Magenta vessels were generally too small to house more than several shoots in a vial, so we changed and placed each vial of shoots into a 3.79-liter plastic jug (Mainstays Canister; Walmart, Bentonville, AR) with a screw-top lid, the latter which was modified so that it had a screened hole (5 cm diameter) for ventilation.

Sweep Samples. A heavy duty beat net with a 38 cm net ring was used to sample for adult Asian citrus psyllid. Ten sweep samples were taken and the number of adult Asian citrus psyllid per sample was counted. A single sweep sample consisted of five subsweeps (each subsweep an arc varying in length depending on the particular planting, with rough estimates of arc length being 77 cm at Gardens; 90 cm at Citrus; 44 cm at Jupiter; 95 cm at Laurel; and 100 cm at Kings). The differences in arc length confound direct comparisons of sweep counts among the five plantings. The sweep samples were taken along the upper horizontal surface of a hedge, with the net ring in a vertical position and the lower edge of the ring in contact with foliage (depending on hedge architecture and how recently a hedge had been pruned, the contact width of each arc was estimated to range from 20 to 30 cm disturbing foliage to a depth of 3–7 cm). The sweep samples were taken at 10 locations spanning from one end of a hedge to the other. All sweep samples were taken by the same technician between 10 a.m. and noon.

Air Temperatures and Rainfall. Weather data during the study were obtained from three weather stations monitored by Weather Underground (www.wunderground.com, accessed 9 March 2015; see Supp Information 2 [online only]).

Statistical Analyses. Simple means and standard errors of the mean (SEM) were the primary statistics used to describe the data. Population trends of Asian citrus psyllid over time were investigated for each planting by subjecting the following variables to a linear regression over time: mean number of adult Asian citrus psyllid per sweep sample per sample date ($\log_{10}$-transormed); mean density rating for eggs or nymphs per flush shoot per sample date; percentage of shoots infested by eggs or nymphs on each sample date; and mean percentage parasitism per sample date. The linear regressions were conducted using PROC GLM (SAS Institute, 2010) with time being the day of the study (day one being March 7, 2011). Percentage data were arcsine-transformed (Gomez and Gomez 1984) for the regressions.

Results

Air Temperatures and Rainfall. An overview of air temperatures and rainfall in the vicinity of the plantings is presented in Figure 1. Additional information is provided in Supp Information 2 (online only).

Flush Abundance. Flush appropriate for oviposition was almost continually present at each planting (Fig. 2). Flush was consistently most abundant at Gardens and Kings—over all sample dates there was a mean ± SEM of 13 ± 2 shoots per sample at each of these sites, equivalent to 187 shoots per m^2. Fewer flush shoots were generally observed at Citrus and Laurel (over all sample dates, 8 ± 1 and 7 ± 1 shoots per sample, respectively, ~108 shoots per m^2). The Jupiter hedge consistently had the fewest shoots, 4 ± 1 shoots per sample (57 shoots per m^2).

Sweep Samples. Adult Asian citrus psyllid were observed in sweep samples on every sample date at each planting (Fig. 3). A mean of 14, 4, 2, 8, and 55 adults per sweep sample (with SEMs of 2, 1, 0.3, 8, and 18, respectively) was observed over all sample dates at Gardens, Citrus, Jupiter, Laurel, and Kings, respectively. The large mean observed at Kings was the result of a number of large sample peaks, notably 442 ± 86 adults per sample during August 2011, 307 ± 125 during September 2011, and 368 ± 44 during August 2012. At Gardens where no parasitoid releases were made, the mean number of adults per sweep sample was frequently higher than 15 with two prominent peaks, one during August 2011 at 58 ± 9 per sample and one during September 2012 at 47 ± 7 per sample. These were also periods of time that peak outbreaks of adults occurred at the Citrus, Laurel, and Kings plantings. Peak adult outbreaks also occurred at these latter three plantings during late summer 2013. Numbers of adult Asian citrus psyllid per sweep sample were consistently small at Jupiter, never reaching a mean of 6 per sample. For each planting, linear

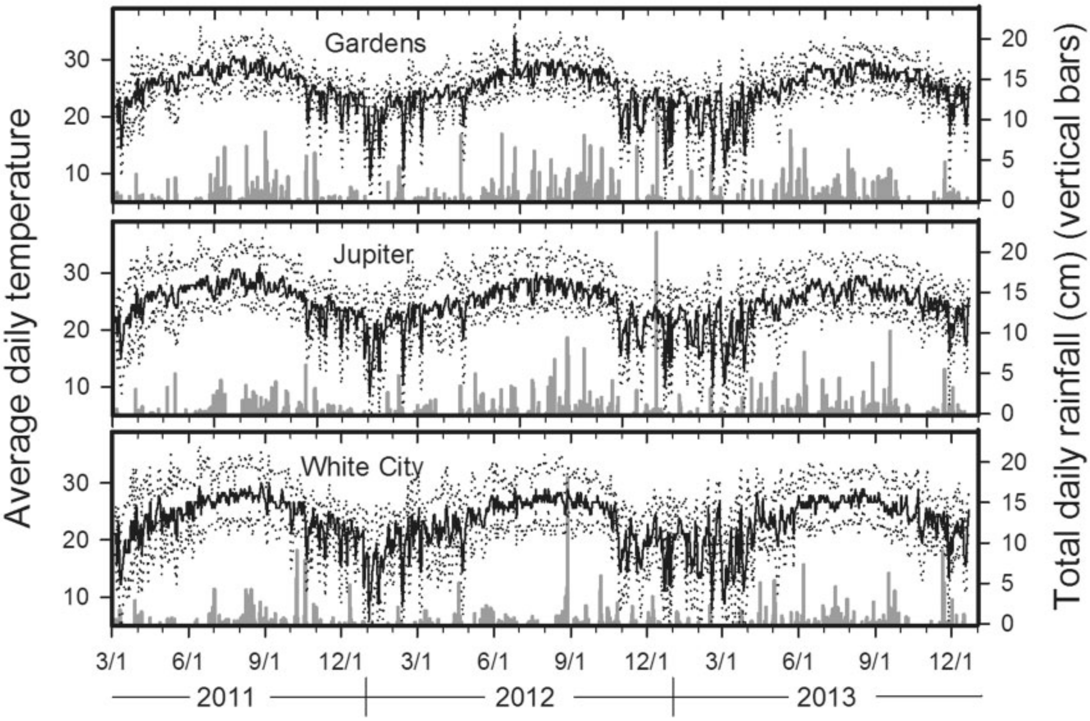

Fig. 1. Daily air temperatures (dark line reflects averages, single data points reflect minimums and maximums) and rainfall (vertical bars) in the vicinity of five urban plantings of orange jasmine in southeast central Florida (Supp Information 1 [online only]). The White City weather station was centrally located among the Citrus, Laurel, and Kings plantings.

regression of the mean number of adults per sweep sample (log-transformed to compensate for the sample extremes at Kings) over time indicated there was no significant change in adult population levels per sweep sample over the 34-mo study (consult trend lines in Fig. 3)—for Gardens, $F_{1,30} = 0.9$, $P = 0.36$; for Citrus, $F_{1,32} = 2.3$, $P = 0.14$; for Jupiter, $F_{1,30} = 3.6$, $P = 0.07$; for Laurel, $F_{1,32} = 0.0$, $P = 0.95$; and for Kings, $F_{1,32} = 0.3$, $P = 0.58$.

Abundance of Eggs and Nymphs. Egg and nymph populations were persistent throughout the study at each planting (Fig. 4). Over all sample dates, eggs and nymphs were consistently most abundant at Kings (mean abundance ratings per shoot of 1.0 ± 0.1 and 0.8 ± 0.1, respectively). Immatures were consistently least abundant at Jupiter (average egg and nymph ratings per shoot of 0.4 ± 0.1 and 0.5 ± 0.1, respectively). Mean egg abundance ratings per shoot at the Jupiter planting declined over time ($F_{1,29} = 6.0$, $P = 0.02$, regression slope $= -0.0004$). For egg ratings at the other four plantings, there was no significant change in egg densities per shoot over time (consult trend lines in Fig. 4)—for Gardens, $F_{1,30} = 0.04$, $P = 0.84$; for Citrus, $F_{1,31} = 2.7$, $P = 0.11$; for Laurel, $F_{1,31} = 2.2$, $P = 0.15$; and for Kings, $F_{1,31} = 0.3$, $P = 0.57$. With respect to nymph density ratings, there was no significant increase or decrease over time at Gardens ($F_{1,30} = 2.6$, $P = 0.12$); Citrus ($F_{1,31} = 0.8$, $P = 0.38$); Jupiter ($F_{1,29} = 3.0$, $P = 0.09$); Laurel ($F_{1,31} = 0.8$, $P = 0.39$); or Kings ($F_{1,31} = 1.6$, $P = 0.21$).

On the average over all five plantings and sample dates, $39.3 \pm 1.8\%$ shoots were observed to be infested by eggs and 46.4 ± 2.0 % shoots were observed to be infested by nymphs. Percentages of shoots infested by eggs and nymphs were consistently highest at Gardens (44 ± 4 and $58 \pm 4\%$, respectively) and Kings (55 ± 5 and $50 \pm 5\%$, respectively); and generally intermediate at Citrus (37 ± 3 and $43 \pm 4\%$, respectively), Laurel (33 ± 3 and $44 \pm 4\%$, respectively) and Jupiter ($37 \pm 3\%$ for each life stage). At Jupiter, a significant decline occurred over time in the percentage of flush shoots infested by eggs ($F_{1,30} = 4.6$, $P = 0.04$, regression slope $= -0.015$). For each of the other four plantings, there was no significant increase or decrease over time in percentages of shoots infested by eggs—Gardens $F_{1,30} = 0.1$, $P = 0.72$; Citrus $F_{1,31} = 2.2$, $P = 0.15$; Laurel $F_{1,31} = 3.4$, $P = 0.08$; and Kings $F_{1,31} = 0.9$, $P = 0.35$. Also for each planting, there was no significant change over time in percentages of shoots infested by nymphs—Gardens $F_{1,30} = 1.9$, $P = 0.17$; Citrus $F_{1,31} = 0.0$, $P = 0.94$; Jupiter $F_{1,30} = 3.7$, $P = 0.07$; Laurel $F_{1,31} = 1.0$, $P = 0.34$; and Kings $F_{1,31} = 0.6$, $P = 0.45$.

Parasitism by *T. radiata*. *T. radiata* was observed throughout the 34-mo study at each of the five plantings (Fig. 5). Based on numbers of adult Asian citrus psyllid and *T. radiata* emerging in the lab, over all sample dates and flush shoots observed there were averages of 2.8, 2.0, 2.4, 3.2, and 2.8 Asian citrus psyllid late instar nymphs per shoot on shoots collected from Gardens, Citrus, Jupiter, Laurel, and Kings, respectively, with

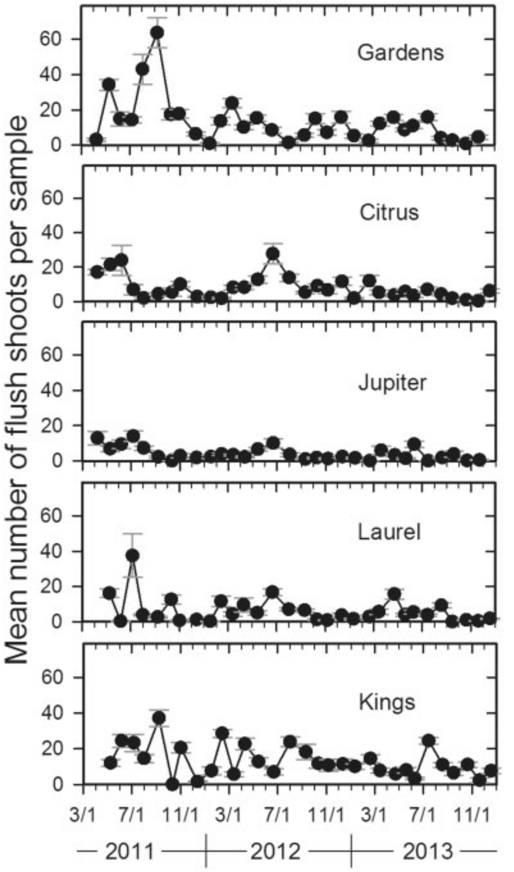

Fig. 2. Abundance of flush (mean number ± SEM per $0.07\,m^2$) associated with orange jasmine at five urban plantings in southeast central Florida.

maximum averages per sample date of 9.5, 5.0, 9.5, 12.6, and 16.0 per shoot, respectively. The single largest number of late instar nymphs observed per shoot was 12 at Gardens, 16 at Citrus, 20 at Jupiter, 9 at Laurel, and 35 at Kings. At the beginning of the study but prior to initiating releases of *T. radiata* at Citrus, Jupiter, and Laurel, relatively low percentages of parasitism were observed at these locations. However, preliminary surveys during 2010 at these study sites indicated percent parasitism was variable but sometimes exceeded 70% (data not presented). Percent parasitism per shoot averaged 35 ± 5% at Gardens; 36 ± 4% at Citrus; 16 ± 4% at Jupiter; 20 ± 4% at Laurel; and 24 ± 5% at Kings. Mean percent parasitism of Asian citrus psyllid did not vary significantly over time at any planting: Gardens $F_{1,28} = 0.6$, $P = 0.44$; Citrus $F_{1,24} = 0.4$, $P = 0.54$; Jupiter $F_{1,24} = 0.1$, $P = 0.78$; Laurel $F_{1,26} = 0.1$, $P = 0.79$; and Kings $F_{1,24} = 0.2$, $P = 0.69$.

Discussion

Asian citrus psyllid reproduction is dependent on young flush for both oviposition and nymph development (Husain and Nath 1927). Flush appropriate for Asian citrus psyllid reproduction was consistently present at the orange jasmine plantings largely because each hedge was regularly pruned and environmental conditions remained generally favorable for flush development throughout the study. Orange jasmine hedges are pruned to maintain a neat appearance. Pruning removes the most recent flush, but pruning also promotes the plant to produce another new flush. The abundance and age of flush shoots at any point in time thus varies depending on how recently a hedge has been pruned. Pruning orange jasmine reduces infestations of Asian citrus psyllid by removing flush with eggs and nymphs resulting in insect death, but rapid resurgence of flush growth promotes re-infestation (Chien and Chu 1996). The plantings we studied were pruned on a variable schedule, usually a light pruning every 3–5 wks depending primarily on growing conditions (sometimes longer at the Jupiter planting due to poorer growing conditions)—clippings fell to the ground and were not hauled away. There were times we would visit a hedge to discover it needed pruning and that relatively few flush shoots appropriate for Asian citrus psyllid immatures remained. There were also times we would visit a hedge that had just been pruned and there had not been enough time for new flush to begin emerging. In either case, we were almost always successful in finding 25 young shoots for the egg and nymph density assessments. However, it was sometimes impossible to find flush shoots infested by older nymphs for assessments of parasitism by *T. radiata*. Future studies on population dynamics of Asian citrus psyllid and its natural enemies associated with orange jasmine might benefit by coordinating pruning and sampling activities as well as examining site-specific weather data and irrigation schedules.

Population levels of Asian citrus psyllid at each planting varied over the 34-mo study, but Asian citrus psyllid was generally most abundant during late summer and least abundant during the winter. The relative abundance of Asian citrus psyllid varied among the five plantings according to the general health of each hedge, with the highest population levels associated with the Kings hedge which was regularly watered and fully exposed to sunlight while the lowest levels were associated with the Jupiter hedge which was not irrigated and greatly shaded by trees. *T. radiata* attacks nymphs thus the impact of the parasitoid should be reflected in numbers of nymphs and percentages of nymphs parasitized, but ultimately it is numbers of adults that citrus growers strive to reduce. Sweep samples showed that adult Asian citrus psyllid were frequently abundant, particularly at Kings where hundreds of adults were sometimes present in a single sweep sample. Mean abundance ratings for numbers of visible eggs and nymphs on shoots at Gardens and Kings reflected averages of 10 eggs (range 1–20) and 5 nymphs (range 1–10) per shoot, but larger densities of immatures per shoot were frequently present. Actual numbers of eggs and nymphs per shoot would be larger because we only rated the numbers we could see—large numbers of immature Asian citrus psyllid on a shoot can be hidden in plant crevices and between

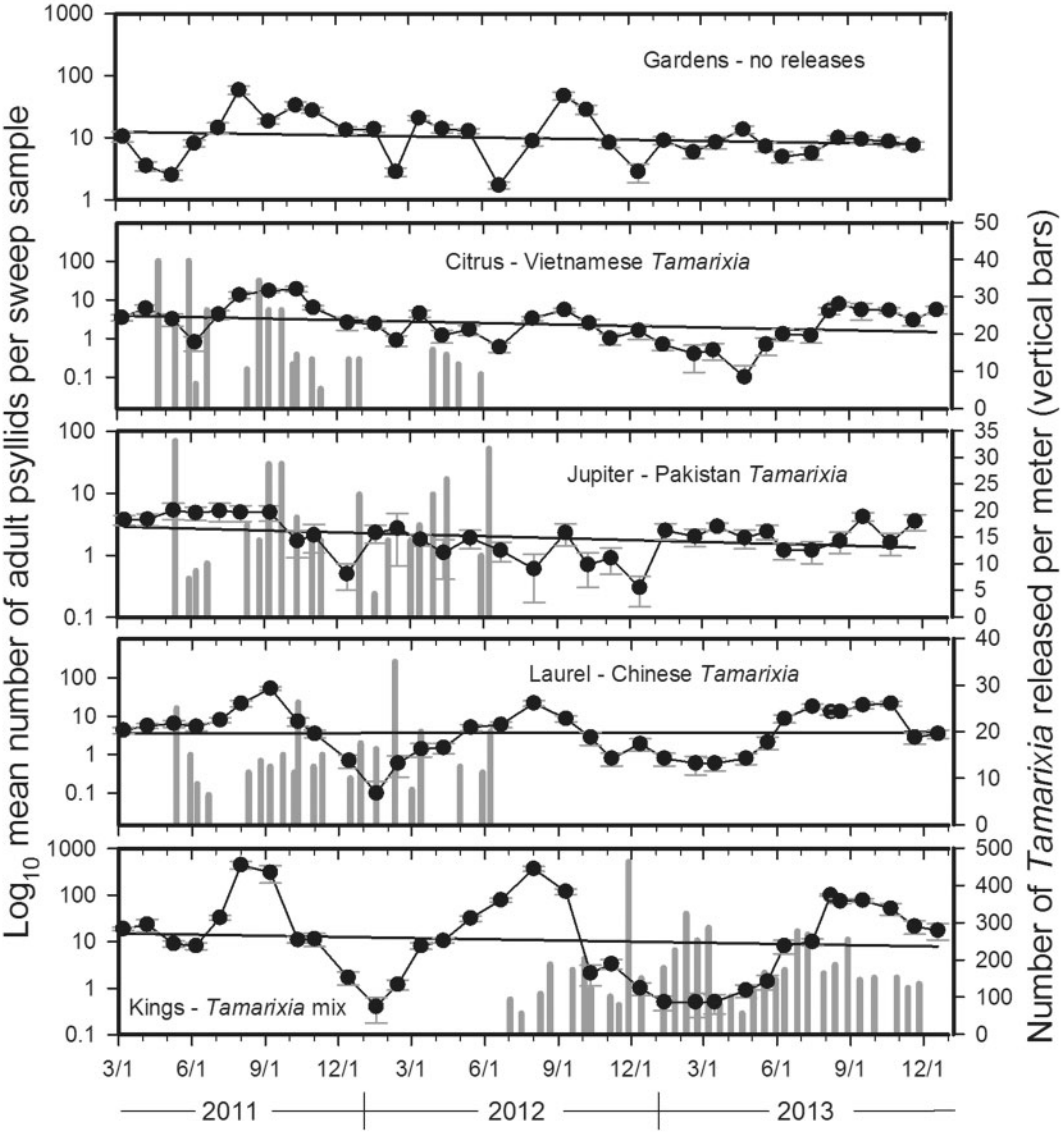

Fig. 3. Mean number (log-transformed) of adult Asian citrus psyllid per sweep sample (black data points) and numbers of adult *T. radiata* released (vertical bars) at five urban plantings of orange jasmine in southeast central Florida. For each planting, the straight black line reflects the temporal trend of numbers of adult psyllids per sweep sample. Existing populations of *T. radiata* at the five plantings were augmented at four plantings by releasing three new haplotypes of the parasitoid (from Vietnam, Pakistan, and China; all three were released at Kings). Note the differences among the five graphs in the scale of the Y axes.

young developing leaflets. In light of the abundance of flush shoots, absolute numbers of eggs and nymphs present at a planting could have sometimes been in the order of thousands. These findings support concerns that orange jasmine in urban areas may promote higher area-wide populations of Asian citrus psyllid that could emigrate to nearby commercial citrus. In fact, there were instances when large numbers of adult Asian citrus psyllid were observed at a planting on one sample date but not the next, which might be attributed to emigration. Hall et al. (2008) reported a large exodus of adult Asian citrus psyllid from citrus, and it is likely that adults may regularly disperse to and from orange jasmine as has been reported for Asian citrus psyllid in citrus (Hall and Hentz, 2011). However, questions remain about how far adult Asian citrus psyllid may regularly disperse (on their own or wind assisted), thus the risk posed to commercial citrus by orange jasmine as a potential source of Asian citrus psyllid is difficult to assess. A full assessment would benefit by knowing how many urban plantings exist in an area. Our initial survey for urban plantings in Saint Lucie County was

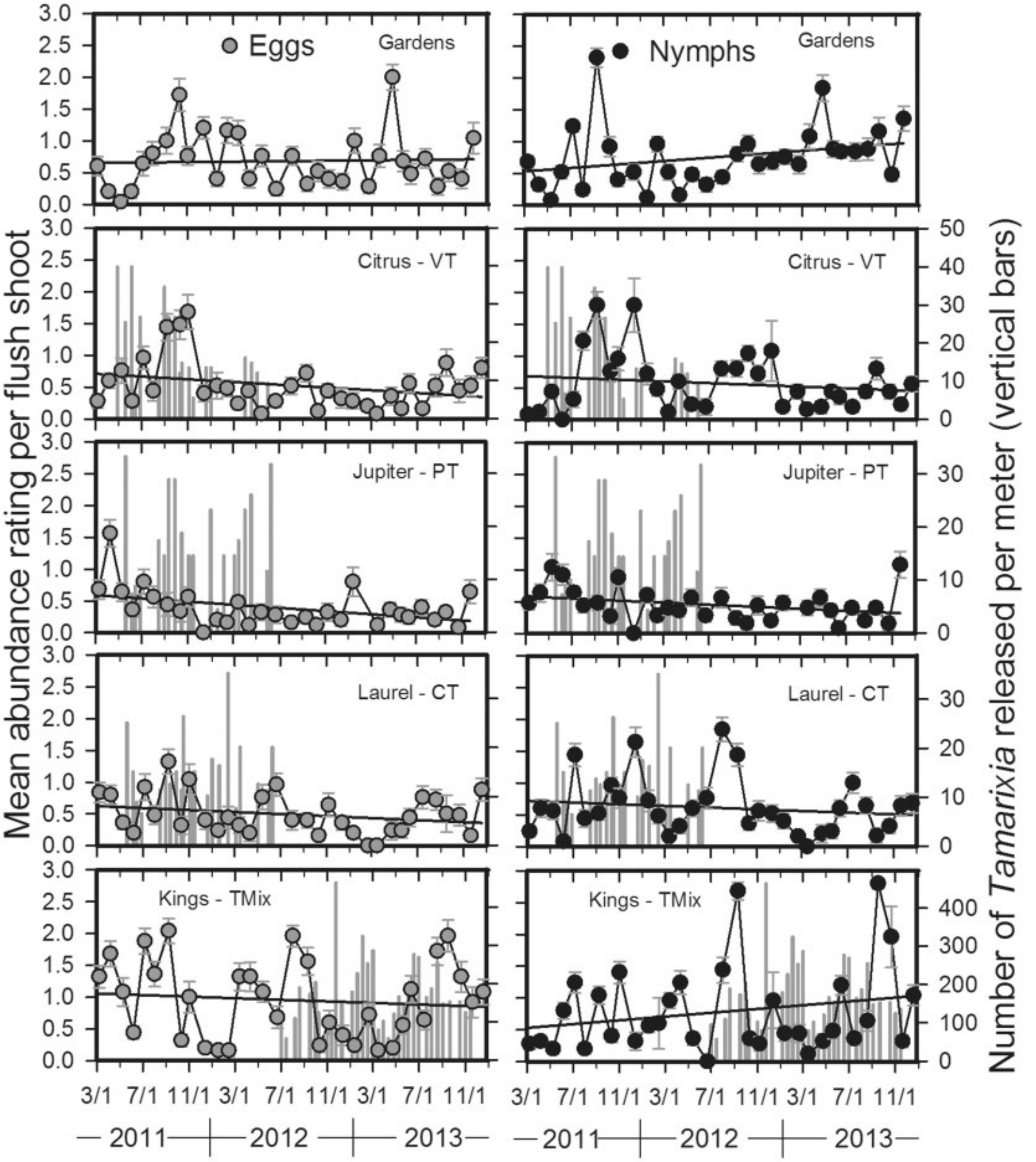

Fig. 4. Mean abundance ratings for densities of eggs and nymphs of the Asian citrus psyllid per flush shoot and numbers of adult *T. radiata* of three new haplotypes released (vertical bars) at five urban plantings of orange jasmine in southeast central Florida. For populations of eggs (solid gray data points) and nymphs (solid black data points) at each planting, the straight black line reflects the temporal trend in abundance ratings. VT, Vietnamese haplotype; PT, Pakistan haplotype; CT, Chinese haplotype. All three haplotypes were released at Kings (TMix).

almost entirely conducted along streets in areas zoned for commercial businesses where we thought large numbers of hedges might be present, and we found seven; we did not survey every street thus more may have been present, and certainly we would have found more had we also searched neighborhoods and gated communities.

Shortly after Asian citrus psyllid was first found in Florida, Tsai et al. (2002) reported that natural enemies did not appear to be key factors in regulating Asian citrus psyllid populations associated with orange jasmine in southern Florida. A later study by Chong et al. (2010) showed Asian citrus psyllid and species of natural enemies known to attack Asian citrus psyllid (including *T. radiata*) were present at each of four urban sites of orange jasmine in south Florida. A 2010 survey conducted of eight urban plantings of jasmine in southeast central Florida (including our Gardens, Citrus, Laurel, and Kings plantings) revealed that the psyllid and parasitoid were present at each site (D.G.H.,

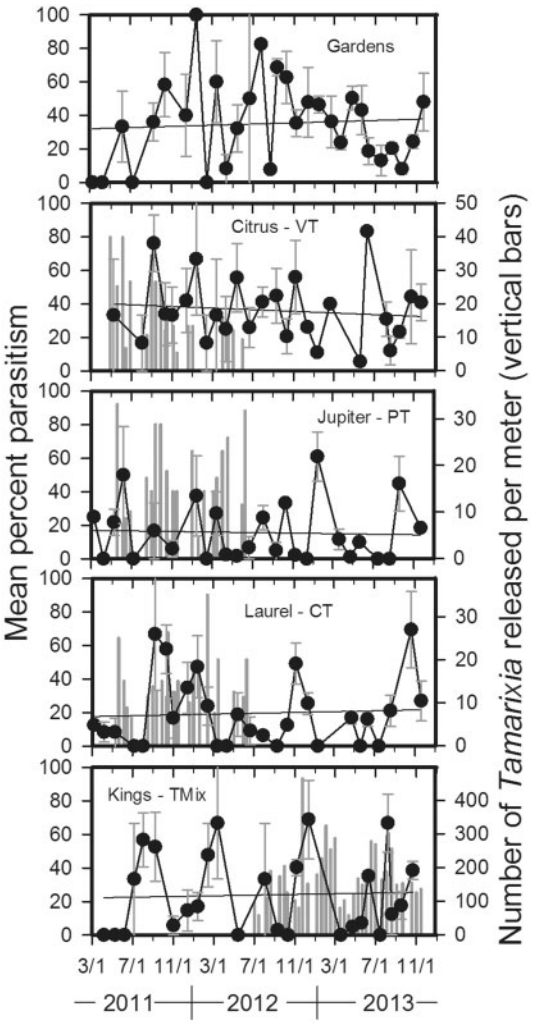

Fig. 5. Percentage parasitism of Asian citrus psyllid by its parasitoid *T. radiata* and numbers of adult *T. radiata* of three new haplotypes released (vertical bars) at five urban plantings of orange jasmine. For each planting, the straight black line reflects the temporal trend of percent parasitism. VT, Vietnamese haplotype; PT, Pakistan haplotype; CT, Chinese haplotype. All three haplotypes were released at Kings (TMix).

unpublished data). Biological control of Asian citrus psyllid by *T. radiata* and other beneficial insects is largely negated in commercial citrus due to current intensive insecticide programs used against Asian citrus psyllid. However, orange jasmine plantings are usually managed without insecticides and thus provide settings in which biological control of Asian citrus psyllid by *T. radiata* and other natural enemies should be promotable. In fact, percentage parasitism of Asian citrus psyllid by *T. radiata* at the five plantings was generally higher and more consistent than in Florida citrus as reported by Hall et al. (2008) and Qureshi et al. (2009). This might be because frequent pruning can enhance *T. radiata* populations by stimulating a constant supply of flush that affords an abundant, continual supply of older nymphs for parasitism. However, pruning orange jasmine can be detrimental to *T. radiata* depending on when pruning occurs, as pruning can remove flush with parasitized nymphs resulting in death of the parasitoids (Chu and Chien 1991; unless perhaps adult parasitoids successfully emerge from mummified nymphs on flush clippings on the ground). In addition to *T. radiata*, an array of other biological control agents known to attack Asian citrus psyllid were frequently abundant at each planting based on sweep samples and visual observations including coccinellids (*Curinus coeruleus* Mulsant, *Cycloneda sanguinea* L., *Harmonia axyridis* Pallas, and *Olla v-nigrum* Mulsant), assassin bugs (*Zelus longipes* L.), brown lacewings (Hemerobiidae), and the entomopathogenic fungus *Hirsutella citriformis* Speare. Periodic Asian citrus psyllid population outbreaks occurred in spite of the presence of these natural enemies.

Of interest was whether the augmentative releases of *T. radiata* provided any measurable benefits in regulating Asian citrus psyllid populations. One desirable long-term benefit would be increased biological control over time associated with the introduction and establishment of a haplotype superior to the one already established in Florida. Of upmost benefit would be the establishment of a haplotype that would permanently reduce the Asian citrus psyllid population equilibrium level. In the short term, releases of *T. radiata* might be used like a bio-pesticide to greatly reduce population levels of nymphs through host feeding and parasitism. At the release rates and frequencies we used, there was no evidence at Citrus, Jupiter, or Laurel that releases of the parasitoids provided any short- or long-term reductions in Asian citrus psyllid population levels. At the Kings planting where all three haplotypes of the parasitoid were released, there was no evidence that the releases provided any increased regulation of Asian citrus psyllid through to the end of the study. Ironically, percentage parasitism by *T. radiata* was generally highest at the Gardens planting where no parasitoid releases were made. It remained possible that some future benefit could still develop over time from the introduction of one or more of the new haplotypes. However, it is likely that pruning negated the full potential of *T. radiata* against Asian citrus psyllid at these orange jasmine plantings. Chien and Chu (1996) recommended that large scale pruning of orange jasmine be avoided because it creates an unstable situation reducing the effectiveness of *T. radiata* against Asian citrus psyllid. This recommendation is supported by observations in Florida on Asian citrus psyllid infestation levels in orange jasmine only trimmed two or three times annually (D. G. Hall, personal observations).

Future investigations on the impact of *T. radiata* on Asian citrus psyllid populations associated with orange jasmine might benefit from taking actual counts of eggs and nymphs including a closer examination of nymphs on shoots to evaluate instars present and to quantify numbers killed by predation. With respect to pruning orange jasmine, the positive and negative impacts of different pruning schedules on Florida populations of

Asian citrus psyllid and *T. radiata* should be further explored. Future research on managing Asian citrus psyllid populations using releases of *T. radiata* as a biopesticide could investigate higher release rates, and these rates might be based on population densities of eggs or nymphs per flush shoot in conjunction with flush shoot abundance.

Supplementary Data

Supplementary data are available at *Journal of Economic Entomology* online.

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