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Research

Population Ecology

Modeling the Phenology of Asian Citrus Psyllid (Hemiptera: Liviidae) in Urban Southern California: Effects of Environment, Habitat, and Natural Enemies

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Abstract

Modeling can be used to characterize the effects of environmental drivers and biotic factors on the phenology of arthropod pests. From a biological control perspective, population dynamics models may provide insights as to when the most vulnerable pest life stages are available for natural enemies to attack. Analyses presented here used temperature and habitat dependent, instar-specific, discrete models to investigate the population dynamics of Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Liviidae). This pest is the target of a classical biological control program with the parasitoid *Tamarixia radiata* (Waterston) (Hymenoptera: Eulophidae). The population trends of *D. citri* eggs, nymphs, and adults, citrus flush growth patterns, and *T. radiata* activity were monitored monthly on orange and lemon trees at 10 urban sites in southern California for a 2-yr period. Cumulative *D. citri* egg, nymph, and adult days recorded at each site, were regressed against accumulated degree-days (DDs) to model the population dynamics of each development stage in relation to temperature. Using a biofix point of 1 January, the model predicted that 10% and 90% of eggs were laid by 198 and 2,255 DD, respectively. Populations of small and large *D. citri* nymphs increased slowly with 90% of the population recorded by 2,389 and 2,436 DD, respectively. *D. citri* adults were present year round with 10 and 90% of the population recorded by 95 and 2,687 DD, respectively. The potential implications of using DD models for optimizing inoculative releases of natural enemies, such as *T. radiata* into citrus habitat infested with *D. citri*, are discussed.

Key words: biological control, *Diaphorina citri*, Weibull distribution, degree-day, *Citrus*

Characterizing the patterns of pest seasonal activity and abundance is one cornerstone of effective pest management (Pedigo 1999). Temperature mediates insect development and behavior, and the phenology of pestiferous life stages can be estimated by degree-day (DD) accumulation (Cammell and Knight 1992, Nietschke et al. 2007, Jones et al. 2009). DD models have been used to predict the occurrence of many economically important aboveground (Doerr et al. 2005, Jones et al. 2013) and subterranean taxa (Johnson et al. 2007, Milosavljević et al. 2017a). These estimates are used to initiate control programs (especially the timing of management tactics) that target the critical life stages of the pest(s) needing population suppression. From a biological control perspective, phenological models assist with management decisions (e.g., making inoculative or augmentative releases of natural enemies) that focus on the key developmental stages of pest targets with the aim of maximizing the efficacy of biocontrol agents (Broatch et al. 2006, Logan et al. 2006). Furthermore, incorporation of DD models into larger decision support systems can promote natural enemy activity thereby enhancing biological control programs across broad geographic regions (Jones et al. 2010). In decision support systems, biocontrol practitioners may receive targeted forecasts of how pest populations might change over time based on their geographic location and associated climate patterns which drive predictions for pest phenology models. Collectively, this information can guide decision making for inoculative or augmentative releases of natural enemies.

The Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Liviidae), is a serious global pest of citrus (Grafton-Cardwell et al. 2013, Milosavljević et al. 2017b). *D. citri* can directly damage citrus plants by feeding on newly emerging foliage, depleting phloem sap, which decreases growth and quality of young, non-bearing plants (Halbert and Manjunath 2004). More importantly, *D. citri* vectors a bacterium, *Candidatus* Liberibacter asiaticus (CLas), the causative agent of a devastating and incurable citrus disease, huanglongbing (Bové 2006, Gottwald 2010). Together, *D. citri* and CLas, represent a formidable pest complex that can render citrus production unprofitable (Yang et al. 2006, Hall et al. 2013). As CLas spreads rapidly through *D. citri* populations, vector control is critical to retard pathogen spread across citrus bearing landscapes (Boina et al. 2009, Lewis-Rosenblum et al. 2015).

In California (United States), urban-grown citrus constitutes a significant percentage of total citrus plants grown (Hoddle 2012). These residential environments are important reservoirs for *D. citri* and CLas in southern California from which dispersal of *D. citri* to commercial citrus groves occurs (Bayles et al. 2017). Biological control is the primary means of control for *D. citri* in urban areas because management with insecticides is either minimal or not feasible (Hoddle and Pandey 2014). Two host-specific parasitoids, *Tamarixia radiata* (Waterston) (Hymenoptera: Eulophidae) and *Diaphorencyrtus aligarhensis* (Shafee, Alam and Argarwal) (Hymenoptera: Encyrtidae) have been introduced and released into California as classical biological control agents targeting *D. citri* (Hoddle et al. 2014). As *D. citri* has not yet become widely established in commercial citrus, and the distribution of CLas is largely restricted to residential areas of three counties in southern California (Los Angeles, Orange, and Riverside), the two parasitoid species are being released almost exclusively in urban areas where the *D. citri*–CLas complex primarily resides (Milosavljević et al. 2017b). To date, more than 6 million parasitoids (*T. radiata* and *Di. aligarhensis* combined) have been mass-produced and released in southern California by the California Department of Food and Agriculture to control *D. citri*, and by extension, to slow further movement of CLas (CDFA 2017).

One of these parasitoids, *T. radiata*, appears to be important for reducing *D. citri* densities infesting backyard citrus (Kistner et al. 2016a,b), but impacts are variable across locations over time. The efficacy of inoculative releases of mass reared parasitoids of *D. citri* in urban California could be enhanced through an improved understanding of temporal fluctuations in population dynamics and the concomitant relative densities of *D. citri* life stages most vulnerable to attack by parasitoids. Information on pest life stage phenology could be used for more precise targeting of parasitoid releases in different locations at times of year when *D. citri* life stages most susceptible to parasitism (or host feeding) are likely to be present. Development and use of temperature-based models for *D. citri* management in advance of the anticipated widespread establishment of this pest in commercial citrus production areas (e.g., the San Joaquin Valley, California) would provide a tool to assist with the development of sustainable management practices that could rely jointly on natural enemy activity and insecticide applications carefully timed to maximize impact on *D. citri* and minimize mortality of key biocontrol agents.

To develop phenological models for *D. citri*, the seasonal population dynamics of this pest on residential lemon and orange trees in southern California was studied for 2 yr. Specifically, we examined the effects of temperature variation on the population abundance of discrete *D. citri* development stages over time in urban environments. We used these findings to develop phenology models for key *D. citri* life stages or life stage groups based on accumulated heat units. Specifically, densities of *D. citri* eggs, small nymphs (i.e., instars 1–3) and large nymphs (i.e., instars 4–5) that are preferentially parasitized by *D. aligarhensis* and *T. radiata*, respectively, and abundance of adult psyllids were recorded. The effects of relative humidity, host plant phenology, and parasitoid activity on development of immature *D. citri* and subsequent population structure were also assessed. The goal of this work was the development of a tool for modeling *D. citri* population phenology for assisting decisions pertaining to ongoing parasitoid releases so as to maximize the likelihood of establishment in areas with *D. citri* infestations.

Materials and Methods

Study Sites

D. citri populations were monitored on citrus in 10 urban sites located in five counties (Riverside, Los Angeles, Orange, San Bernardino, and San Diego) in Southern California from 1 January 2015 to 31 December 2016 (Fig. 1; Supplementary Table 1). Each

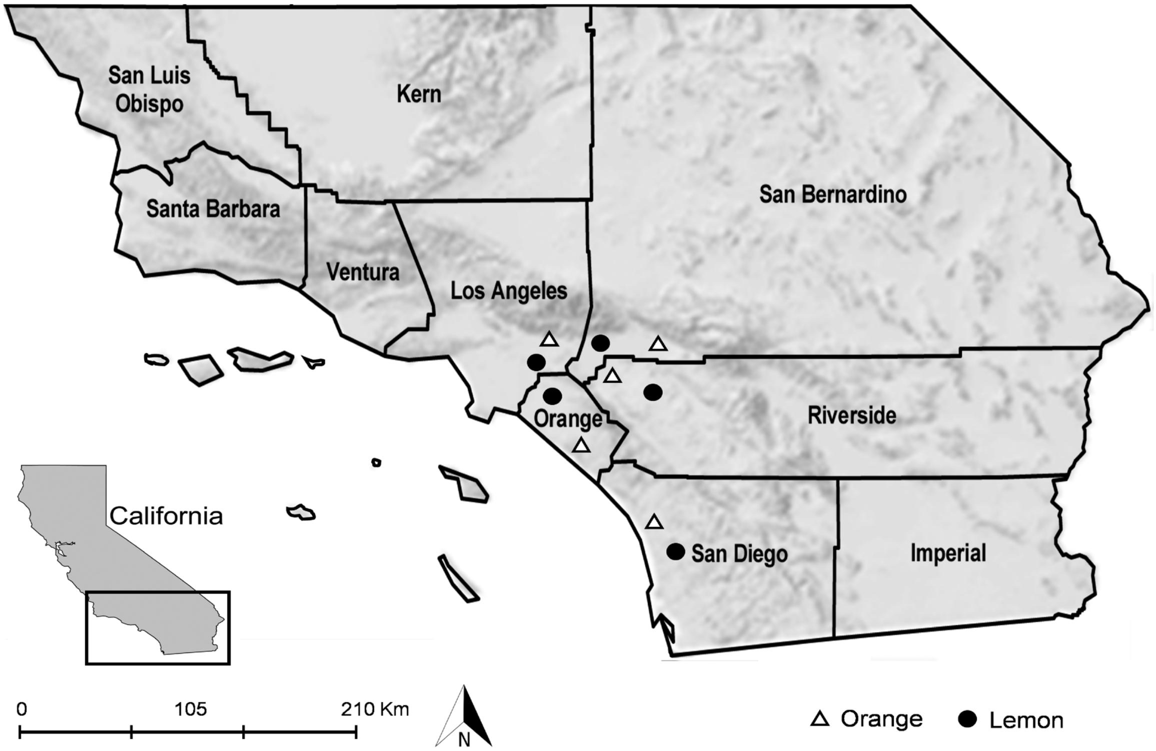

Fig. 1. Map of sites in southern California that were sampled over 1 January 2015 to 31 December 2016 for *D. citri*. The shape of the *symbol* and the *color* denote sampled citrus plant. Each site had either an orange or lemon tree, for a total of 10 study trees that were not treated with insecticides for the duration of this project.

site had either an orange (*Citrus sinensis* [L.] Osbeck) or lemon (*Citrus limon* [L.] Osbeck) tree, (all trees were > 5 yr of age), for a total of 10 study trees that were not treated with insecticides for the duration of this project. The *D. citri* parasitoids, *T. radiata* and *D. aligarhensis*, were not released at these 10 sites as part of the classical biological control program targeting *D. citri*. Each study site was located approximately 2 km from the nearest parasitoid release locations (CDFA 2017). Parasitism of *D. citri* nymphs by *T. radiata* was found at all study sites. *D. aligarhensis* activity was not observed at these locations. All study sites were situated in 280 to 480 mm annual precipitation zones (Supplementary Table 1; CIMIS 2017).

Data Collection

D. citri populations were monitored monthly at each site throughout the 2-yr evaluation period. Each surveyed tree was divided into four cardinal quadrants (north, east, west, and south), that were examined for *D. citri* life stages on each sampling date. At each study site, timed 2 min visual inspections per quadrant (for a total of 8 min per tree) were made and the numbers of observed adult *D. citri* per tree were recorded. These data were used to calculate the cumulative *D. citri* adult load (i.e., insect days per tree) for each sampled tree over time ([see Data analyses]; Ruppel 1983). At each sampling date, air temperatures and relative humidity for each study site were obtained from the closest weather station. Each study site was located approximately 10 km from the nearest meteorological station (Supplementary Fig. 1; CIMIS 2017).

D. citri oviposition, nymphal development, and subsequent population growth are limited by the presence of soft young tissue (i.e., flush) on host trees (Hall et al. 2008). To measure the prevalence of flush on trees, a 74-cm-diameter steel hoop (this circumscribed an area of 172 cm^2) was randomly placed on the outer foliage and the growth stage of foliage (i.e., presence or absence of flush growth) per tree that was enclosed by the hoop was determined and recorded each month for a total of four hoop samples per tree (Kistner et al. 2016a). These data were used to determine the percentage of sampled foliage that was suitable by sampling date and site for oviposition and the development of immature *D. citri*. The percentage of canopy samples bearing flush growth was calculated as the number of canopy samples with flush growth/(number of flush growth canopy samples + non-flush growth canopy samples) × 100 (Khan et al. 2014).

In addition to timed adult counts and measures of flush availability, two flush shoots suitable for oviposition were randomly excised from each quadrant of each surveyed tree to yield eight flush samples per study tree. Each individual flush shoot was placed in a plastic bag and labeled with the site location and sampling date, placed in a cooler, and transported under California Department of Food and Agriculture permit 2870 to the University of California Riverside Insectary & Quarantine Facility. Excised flush was examined under a dissecting microscope and the absolute number of *D. citri* eggs, small nymphs (instars 1–3), and large nymphs (instars 4–5), and the number of nymphs parasitized by *T. radiata* were recorded. Flush length, measured in cm from the base of the growth to the apex of the stem, from which counts were made, was recorded also (Supplementary Fig. 2), (see Kistner et al. 2016a for details). If samples could not be processed on the day of collection they were kept in a refrigerator (~4°C) for a maximum of 2 d before being examined. Egg and nymph counts per cm of flush shoot were used to assess the cumulative *D. citri* egg and instar load for each surveyed tree over time (see Data Analyses; Ruppel 1983). Percentage parasitism (i.e., total number of parasitized large nymphs/[(total number of non-parasitized large nymphs + total number of parasitized large nymphs) × 100]) was recorded for each study site and date.

Data Analyses

To assess the intensity of the cumulative *D. citri* infestation burden on each individual tree over time, average monthly numbers of *D. citri* eggs, small and large nymphs per centimeter of flush shoot, and adult counts, were converted to corresponding insect days:

$$\text{insect days} = \left(X_{i+1} - X_i\right) * \left[\frac{\left(Y_i + Y_{i+1}\right)}{2}\right] \tag{1}$$

where X_i and X_{i+1} are adjacent points of time about 30 d apart, and Y_i and Y_{i+1} are the corresponding *D. citri* counts (Ruppel 1983). These data were used to compute the cumulative population load of each *D. citri* life stage over time for each study tree (Khan et al. 2014).

To analyze whether *D. citri* populations were affected by temperature, relative humidity, host plant phenology, and parasitoid activity over time, repeated measures generalized linear models with year, host plant (orange, lemon), temperature, presence or absence of flush, relative humidity, parasitism rate, sample month, and all the two-way interactions (fixed effects) as explanatory variables were used for analyses. Repeated measurements were the *D. citri* days recorded at each site for each month of the study. *D. citri* count data necessitated that models were fit with a negative binomial distribution (Sileshi 2006, Ver Hoef and Boveng 2007, O'hara and Kotze 2010). Separate models were run for each of the four response variables (counts of *D. citri* eggs, small nymphs, large nymphs, and adults). The original, more complex, models included all of the fixed terms. However, nonsignificant explanatory variables ($P > 0.15$) were removed to obtain the minimal adequate model for each response variable. After models were selected, model selection with information criteria (Akaike's information criterion) was performed to determine whether the model with fewer variables fit the data as well as the full model (Whittingham et al. 2006, Milosavljević et al. 2016). Spearman's rank-correlation analyses were performed to assess the relationship between *D. citri* life stage densities and flush length. All statistical analyses were performed in SAS (version 9.2, SAS Institute 2009).

Cumulative *D. citri* egg, nymph, and adult days (i.e., percentage of total insect-days calculated over each of the two 12-mo sampling periods) were then regressed against accumulated daily heat units to model the population dynamics of each development stage in relation to temperature. DD accumulations, based upon air temperatures at each study location, were calculated for each survey year from a biofix date of January 1 using the direct calculation method (Jones and Brunner 1993):

$$GDH = T_H - T_{base}; \tag{2a}$$

$$\text{if } T_H < T_{base}, \text{ then } GDH = 0; \tag{2b}$$

$$\text{if } T_H > T_{UT}, \text{then } GDH = \left(T_H - T_{base}\right) - \left(T_H - T_{UT}\right); \tag{2c}$$

$$GDD = \frac{\Sigma GDH}{N} \tag{2d}$$

where GDH is a measure of hourly heat units, GDD is a measure of daily heat accumulations, T_H is the hourly air temperature reading at each sampled location and date, N is the number of hourly readings in a 24-hr period, and T_{base} and T_{UT} are lower and upper development thresholds of each *D. citri* life stage, respectively. To calculate hourly heat units, the lower thresholds were subtracted from the hourly temperatures (equation 2a) and negative heat unit values were adjusted to 0 (equation 2b). When the observed hourly temperature was higher than the upper threshold, the difference between the upper threshold and the hourly reading was subtracted from the heat values (equation 2c). DDs were calculated by dividing the sum of hourly heat units between the thresholds by the number of readings in a 24-hr period (equation 2d). For each study site, hourly air temperatures were obtained from the nearest meteorological station (CIMIS 2017). The lower development threshold values (T_{base}) used for *D. citri* eggs, nymphs, and adults were 9, 10, and 11°C, respectively (Liu and Tsai 2000). The upper development threshold values (T_{UT}) used for *D. citri* eggs, nymphs, and adults were 33, 33, and 41°C, respectively (Liu and Tsai 2000, Hall et al. 2011). A biofix point of 1 January was used because it was the earliest date on which flush was observed on oranges and lemons in southern California (Kistner et al. 2016a).

Cumulative *D. citri* load based on accumulated DDs was modeled using a four-parameter Weibull distribution:

$$y = a * \left[1 - e^{-\left| \frac{x - x_{50} + b * ln(2)^{\frac{1}{c}}}{b} \right|^{c}} \right] \qquad (3)$$

where y is the percent cumulative *D. citri* days, x is the observed cumulative DDs, a, b, c, and x_{50} are empirically derived constants, x_{50} is the time point in DDs at which 50% of the monitored population was recorded, a is the theoretical maximum cumulative *D. citri* days, and b and c are scale (denotes the statistical dispersion of the distribution), and shape (denotes how symmetrical the curve is) parameters, respectively (Table 2; [Weibull 1951, Milosavljević et al. 2017a]). Separate, four-parameter Weibull distribution models, were constructed for each *D. citri* life stage using SigmaPlot 12.0 (Systat Software, Inc, San José, CA). Models were fit across all surveyed

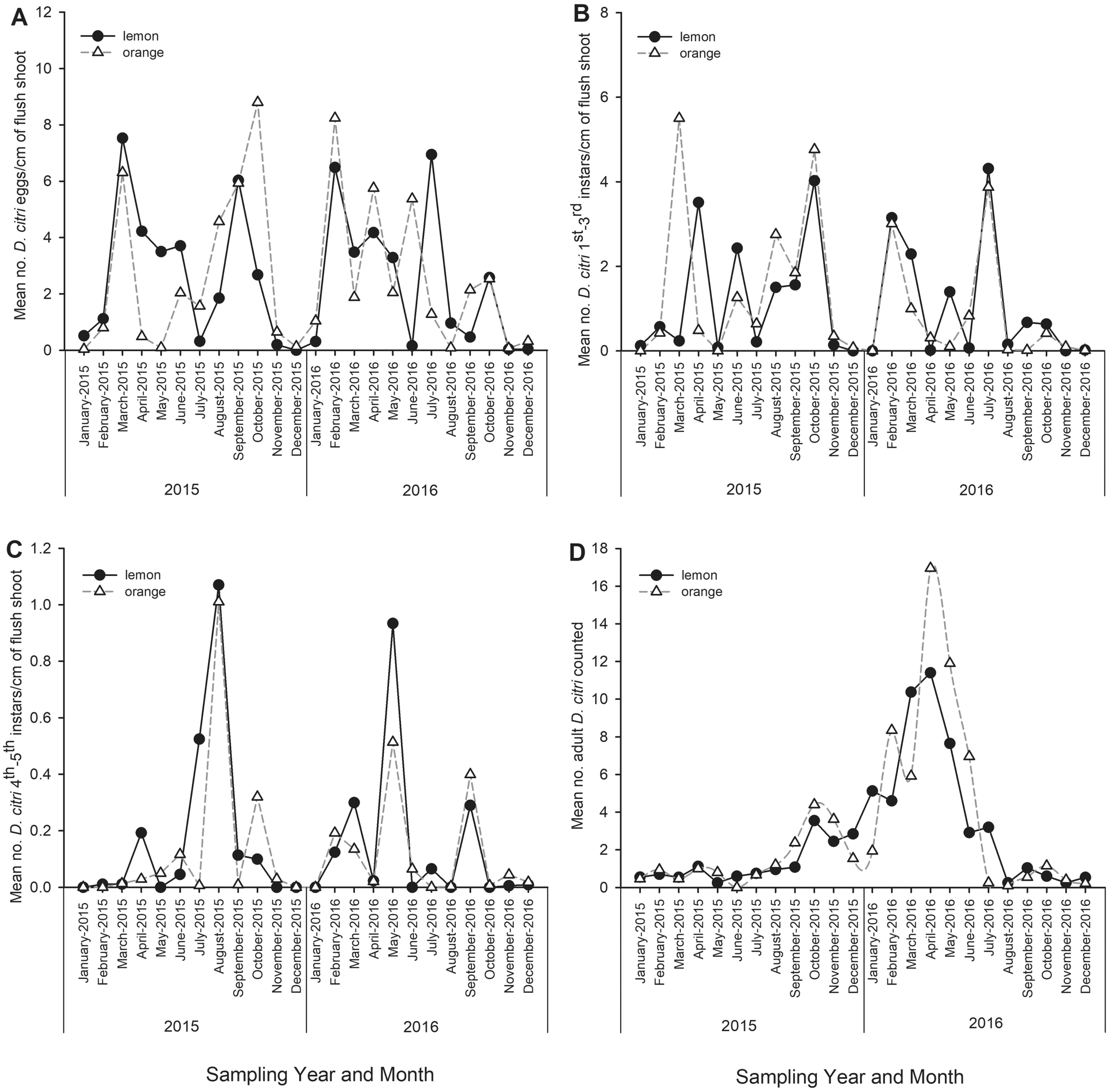

Fig. 2. Relationship between host plants and monthly *D. citri* population densities. Shown are (**A**) mean number of *D. citri* eggs per cm of sampled flush shoot, (**B**) mean number of *D. citri* 1st–3rd instars (i.e., small nymphs) per cm of flush shoot sampled, (**C**) mean number of *D. citri* 4th–5th instars (i.e., large nymphs) per cm of flush shoot sampled, and (**D**) mean number of *D. citri* adult counts (from timed observations), for each host plant (pooled across study sites).

trees as the development of *D. citri* life stages did not differ significantly between oranges and lemons, the two citrus plants used in this study (see Results).

Results

Effects of Abiotic and Biotic Factors on the Seasonal Population Dynamics of *D. Citri*

The average densities of each of the four *D. citri* development stages (i.e., eggs, small and large nymphs, and adults), presence or absence of flush, and rates of parasitism by *T. radiata* over 1 January 2015 to 31 December 2016 are shown in Figs. 2 and 3. *D. citri* eggs were present from January to October, but were mostly absent in November and December (Table 1A month effect; Fig. 2A). The presence and density of *D. citri* eggs were consistently mediated by temperature and citrus flushing patterns (Table 1A; Figs. 2A and 3A), and overall flushing patterns were similar on the two citrus hosts (i.e., oranges and lemons) across seasons (Fig. 4). The number of eggs laid by *D. citri* females was not significantly affected by year, host plant, relative humidity, *T. radiata* parasitism, or interactions between these variables ($P > 0.15$ for all models). Mean monthly egg densities in 2015 and 2016 were positively correlated with average flush length ($r_s = 0.44$; $P < 0.001$).

Similar to eggs, the presence and density of small and large *D. citri* nymphs were significantly affected by temperature and citrus flush growth patterns ("Table 1B and C: $P < 0.04$, for all models; Figs. 2B and C and 3A). Populations of small and large *D. citri* nymphs were abundant from February to October and typically had 1 to 3 peaks per year, around approximately February–March, June–July, and to August–September each year ("Table 1B and C month effects, Fig. 2B and C). Trends in nymph densities over time were similar on oranges and lemons (Figs. 2B and C and 4). Populations of small and

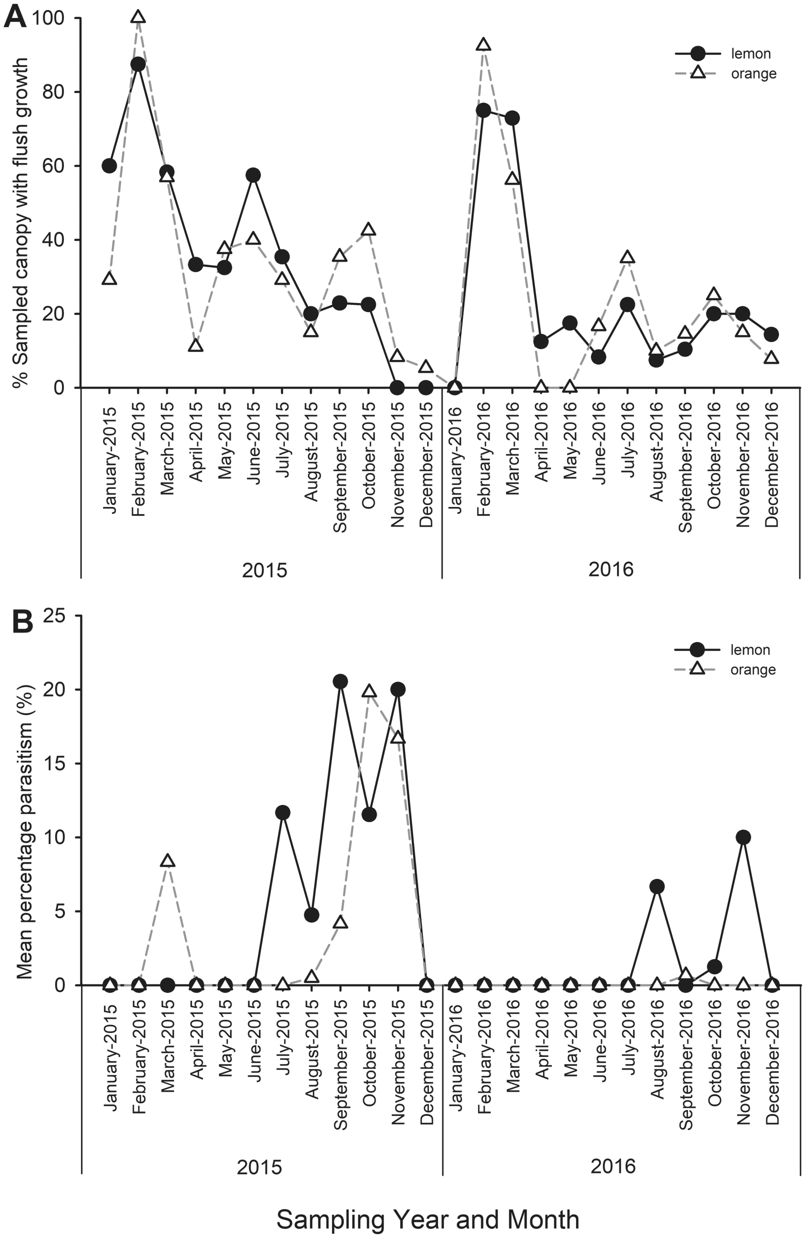

Fig. 3. (**A**) Monthly flushing patterns for oranges and lemons combined across 10 study sites. (**B**) Monthly percentage parasitism trends for *T. radiata* attacking *D. citri* nymphs on oranges and lemons. Parasitism data were combined for each host plant across study sites.

Table 1. Results of repeated measures analyses examining the effects of year, citrus hosts (i.e., orange or lemon), temperature, relative humidity, presence or absence of flush, *T. radiata* parasitism, and month on cumulative *D. citri* days (eggs, small [i.e., 1st–3rd instar] nymphs, large [i.e., 4th–5th instar] nymphs, and adults) recorded across field sites in 2015 and 2016

(A) Eggs	df	χ^2	*P*
Temperature (T)	1	4.40	0.0359†
Flush (F)	1	4.18	0.0408†
Month (M)	11	5.27	0.0217†
T × F	1	4.95	0.0261†
T × M	11	4.77	0.0289†
F × M	11	5.68	0.0172†
(B) Small nymphs	**df**	**χ^2**	***P***
Temperature (T)	1	5.36	0.021†
Flush (F)	1	7.62	0.006‡
Month (M)	11	4.51	0.034†
T × F	1	8.33	0.004‡
T × M	11	4.63	0.031†
F × M	11	4.08	0.043†
(C) Large nymphs	**df**	**χ^2**	***P***
Temperature (T)	1	13.74	0.0002‡
Flush (F)	1	12.79	0.0003‡
Parasitism (P)	1	2.79	0.095*
Month (M)	11	4.10	0.0429†
T × F	1	11.25	0.0008‡
T × M	11	3.77	0.041†
F × M	11	4.15	0.042†
P × M	11	4.41	0.036†
(D) Adults	**df**	**χ^2**	***P***
Year (Y)	1	6.68	0.0098‡
Temperature (T)	1	6.06	0.014†
Month (M)	11	7.34	0.007‡
Y × M	11	6.38	0.011†
T × M	11	3.85	0.041†

Only minimal adequate models for each response variable are shown.
$^{*}P < 0.1$; $^{\dagger}P < 0.05$; $^{\ddagger}P < 0.01$.

large nymphs were not affected by year, host plant species, or relative humidity, or interactions between these variables ($P > 0.16$ for all models). *T. radiata* decreased the survival of large *D. citri* nymphs but this effect was strongly mediated by time. Percentage parasitism peaked from October to December each year (Table 1C parasitism × month interaction effect; Fig. 3B). Population trends of small *D. citri* nymphs were not affected by *T. radiata* activity across seasons ($P > 0.18$ for all models). Mean monthly first-third and fourth-fifth instar densities of *D. citri* in 2015 and 2016 were positively correlated with average flush length (first–third instars: $r_s = 0.61$; $P < 0.001$; fourth–fifth instars: $r_s = 0.67$; $P < 0.001$).

In contrast to eggs and nymphs, densities of *D. citri* adults varied substantially across study years (Table 1D; Fig. 2D). Significantly more adults were recorded in 2016 than 2015 (Fig. 2D), but overall densities of adult *D. citri* were similar on oranges and lemons (Fig. 2D). The presence and density of *D. citri* adults were consistently mediated by temperature patterns (Table 1D). Population levels of *D. citri* adults were not affected by flush patterns, relative humidity, or *T. radiata* parasitism, or any interactions with these variables ($P > 0.15$ for all models). No significant correlation was found between 2015 and 2016 mean monthly adult densities and average flush length ($P > 0.05$).

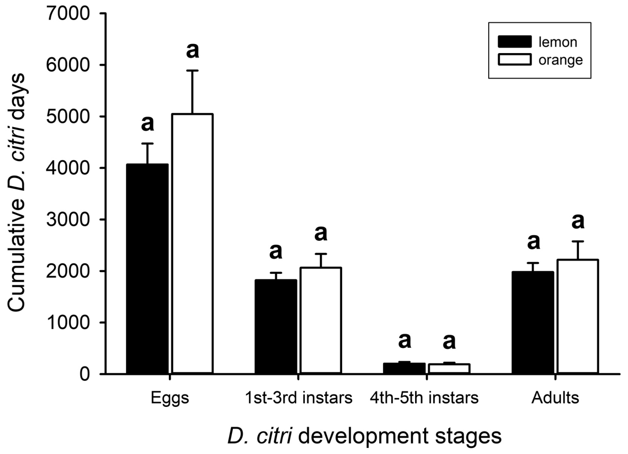

Fig. 4. Mean (±SE; untransformed) cumulative *D. citri* egg, 1st–3rd instar (i.e., small nymphs), 4th–5th instar (i.e., large nymphs), and adult days from 2015 to 2016 on oranges and lemons. Different letters above bars within a life stage indicate significant differences between host plants ($\alpha = 0.05$). Cumulative *D. citri* days for each host plant were pooled across study sites.

Phenology Models

Cumulative *D. citri* egg, nymph, and adult days were paired with accumulated DDs for all sampled sites (Fig. 5). Temperature and time constantly mediated the seasonal dynamics of each pest stage. Weibull models described 86, 89, 90, and 82% of the variation of cumulative *D. citri* egg, small nymph, large nymph, and adult densities, respectively (Table 2). The first *D. citri* eggs were recorded early in the year, between 0 and 15 accumulated DDs (1% of the total eggs found each year) from the 1 January biofix point, with 10, 50, and 90% of the total population predicted at 198 (from 2 February to 13 March), 961 (3 May to 13 June), and 2,255 (16 August to 21 September) DDs, respectively (Figs. 5A and 6A). The appearance of the first generation of small nymphs began early in the year at 19 DDs (1% of the total small nymphs found each year), with 10% of the total population predicted at 211 (from 6 February to 23 March) DDs, 50% at 1,015 (30 May to 4 July) DDs, and 90% at 2,389 (9 September to 13 October) DDs from the 1 January biofix point (Figs. 5B and 6B). The predicted appearance of the first generation of small nymphs was well correlated to a backward estimation of observed first egg hatch in the models used for these analyses (Fig. 5A and B). As expected, large nymphs were present in relatively low numbers early in the season (1% of the total population found each year predicted at 250 DDs [beginning of March]), and did not dramatically increase until 368 (from 13 March to 1 May) DDs (10% of the total population found each year), with 50 and 90% of the total annual population predicted at 1,046 (18 June to 25 July) and 2,436 (30 September to 26 October) DDs, respectively from the 1 January biofix point (Figs. 5C and 6C). *D. citri* adults were present either on oranges or lemons throughout the entirety of the 2-yr study period. From the 1 January biofix point, first adults were found between 0 and 10 accumulated DDs (1% of the total population found each year), 10% of the adult population was recorded by 95 (from 16 January to 19 February) DDs, 50% by 912 (18 May to 26 June) DDs, and 90% by 2,687 (28 October to 4 December) DDs (Figs. 5D and 6D).

Discussion

Work presented here demonstrates that the cumulative temporal populations of *D. citri* life stages infesting urban lemon and orange trees in southern California can be predicted using deterministic DD

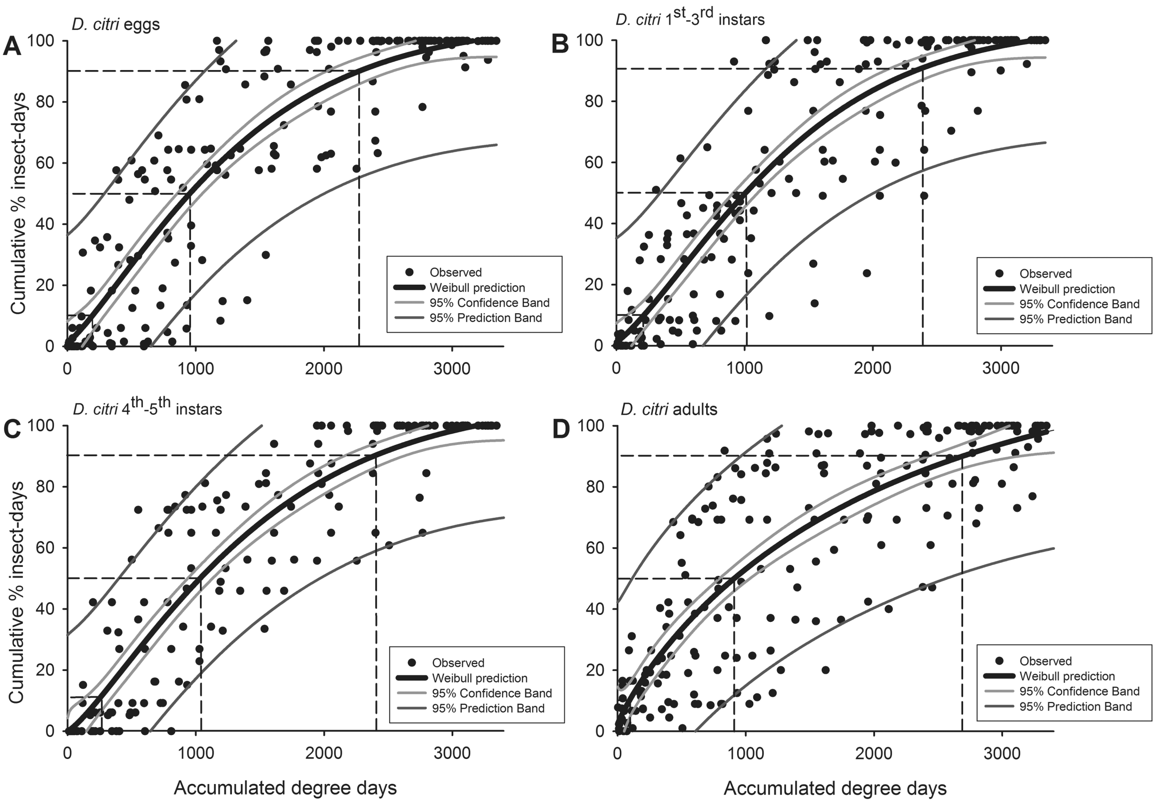

Fig. 5. Cumulative insect-days for each *D. citri* development stage in relation to cumulative degree-days (DDs). Shown are cumulative insect-days of *D. citri* (**A**) eggs (**B**) 1st–3rd instar nymphs (i.e., small nymphs) (**C**) 4th–5th instar nymphs (i.e., large nymphs) and (**D**) adults. The *symbols* represent the raw data, and the curves show the best-fit Weibull model for each *D. citri* life stage with the 95% confidence and prediction bands. The *dashed lines* indicate the number of cumulative DDs where 10, 50 and 90% of the total population was found.

Table 2. The best-fit parameters of the Weibull model for each *D. citri* developmental stage (eggs, small nymphs [i.e., 1st–3rd instars], large nymphs [i.e., 4th–5th instars], and adults)

D. citri life stage	Weibull parameters					
	a	b	c	x_{50}	R^2	Adjusted R^2
Eggs	105.83	1407.49	1.29	1017.16	0.89	0.86
Small nymphs	105.72	1471.93	1.31	1072.95	0.90	0.89
Large nymphs	113.24	1350.31	0.99	1184.73	0.92	0.90
Adults	124.66	2027.73	0.86	1301.22	0.85	0.82

models. Specifically, we examined the relationship between monthly densities of key *D. citri* life stages, host plant species (i.e., lemons and oranges), and cumulative DDs in an effort to predict the abundance of each pest life stage relative to its other life stages across the course of a year. From a pest management perspective, model predictions may assist with pest management decisions (e.g., inoculative or augmentative releases of parasitoids or pesticide treatments) targeting *D. citri* in residential areas in southern California. Our results support those from other studies confirming that temperature and flush growth are fundamental indicators that are positively correlated with densities of all *D. citri* life stages (Liu and Tsai 2000, Tsai et al. 2002, Hall et al. 2008, Kistner et al. 2016a, Udell et al. 2017). Accordingly, the DDs models used here explained over 82% of the variation in cumulative population trends of each *D. citri* life stage. Because these are the first DD models to address the field biology of *D. citri* in southern California, this work provides a foundational framework that could be used for development of web-based decision-support systems for *D. citri* management by citrus growers (and potentially urban-based horticulturalists) in California.

Our results indicated that in southern California, urban-grown oranges and lemons are equally preferred host plants by *D. citri* with similar phenologies and densities of eggs, nymphs, and adults being observed across sites and years. This is in line with previous studies that showed the development of *D. citri* life stages and population trends do not differ significantly between the two citrus species (i.e., oranges and lemons) used in this study (Tsai and Liu 2000, Nava et al. 2007, Kistner et al. 2016a). However, as seen in earlier studies, not all citrus species are equally suitable for *D. citri* across time (Patt and Setamou 2010, Robbins et al. 2012, Khan et al. 2014, Kistner et al. 2016a). Previous work found that limes (*Citrus* × *aurantiifolia* Swingle) and curry leaf plants (*Bergera koenigii* [L.] Sprengel [Rutaceae]) consistently supported higher *D. citri* densities compared to oranges, lemons, and grapefruit (*Citrus* × *paradisi* Macfadyen) (Kistner et al. 2016a). For example, vigorous

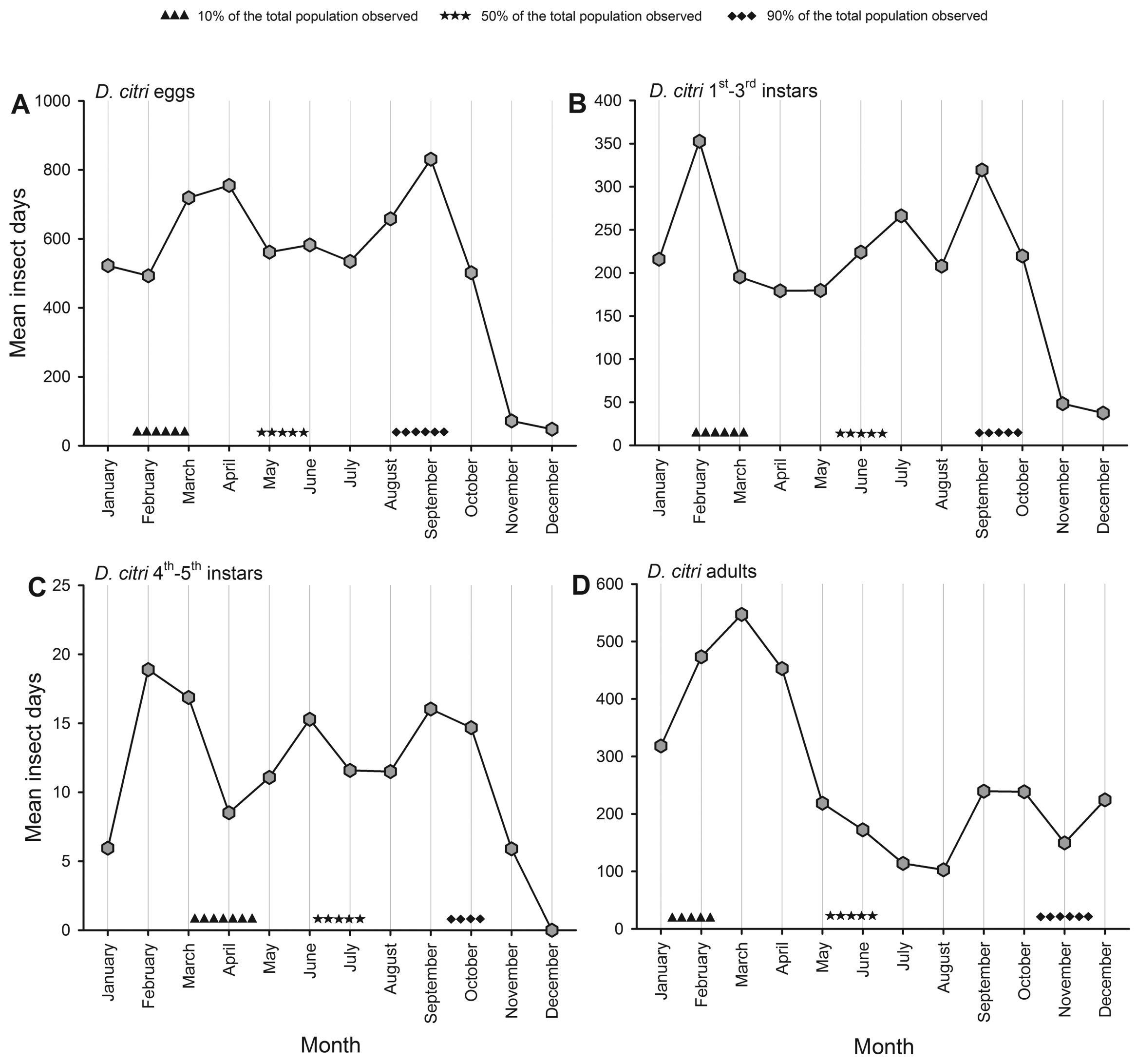

Fig. 6. Line graph showing monthly means of cumulative insect-days of *D. citri* (**A**) eggs (**B**) 1st–3rd instar nymphs (**C**) 4th–5th instar nymphs and (**D**) adults (pooled across 10 study sites). *Symbols* below line graphs denote time periods during which 10, 50, and 90% of the total target population was recorded, for each *D. citri* life stage.

flush growth of lime plants makes them more habitable for *D. citri* eggs and nymphs (Kistner et al. 2016a), while the combination of continuous shoot flushes and olfactory cues produced by curry leaf plants attracts *D. citri* adults (Patt and Setamou 2010). Variation in host plant suitability for oviposition and nymph development and attractiveness to adult *D. citri* suggests that DD models for *D. citri* populations should be developed for pest populations infesting specific host plants.

In addition to temperature, cumulative temporal *D. citri* populations (i.e., eggs and nymphs) in residential areas of southern California consistently coincided with orange and lemon leaf flushing patterns over time. Other studies have shown similar correlations as the seasonal availability of young plant tissue on citrus host trees strongly influences *D. citri* oviposition, nymphal development, and subsequent population growth (Hall et al. 2008, Kistner et al. 2016a). In this study, we tracked specific *D. citri* developmental stages as a function of cumulative egg and nymph densities per flush shoot and temperature combined. Moreover, citrus flush was present year round on both host plant species in 2015 and 2016 but peaked early in the season (January–March) when temperatures were generally lower, justifying the biofix point of 1 January used in these models. Thus, the models developed here could suggest a time when a psyllid life stage should be present and likely abundant, signaling growers to verify that flush and psyllids are abundant before making a management decision.

The application of deterministic DD models to predicting *D. citri* population dynamics is an important initial step for risk assessment, due to minimal assumptions and relative ease of analysis compared to modeling time series growth rates (such as a Ricker [Ricker 1954] or Beverton-Holt models [Clark et al. 1973]). Successfully using DDs to predict cumulative temporal *D. citri* densities, as demonstrated here (adjusted $R^2 > 0.8$) supports the utility of this approach. Urban citrus grown in southern California may, however, support greater year round *D. citri* densities compared to commercial citrus because of the heterogeneity of habitats, diverse plantings in small areas (e.g., oranges, lemons, limes, grapefruit, and kumquats were observed in some gardens), and more intensive management practices (i.e., pruning, watering, fertilization, and minimal pesticide use) by homeowners (Grafton-Cardwell et al. 2013, Kistner et al. 2016a, Bayles et al. 2017). Further, in California, commercial citrus production areas span a diverse variety of climatic zones, hot desert interior regions, moderate intermediate zones, and cooler coastal areas. DD methods, such as those used here, may be readily extended to account for the complex spatio-temporal dynamics across commercial citrus landscapes. However, because these are fundamentally different areas, and the biology and development of *D. citri* is closely tied to flush phenology and the temporal population abundance is strongly influenced by temperature (Thomas et al. 2017), studies similar to those conducted here will be needed.

Unlike other approaches that tracked discrete pest generations (Doerr et al. 2005, Jones et al. 2005, Joshi et al. 2016) or temporal patterns of life-cycle events between two populations (Murtaugh et al. 2012), we monitored discrete *D. citri* life stages, including those most susceptible to parasitism and host feeding. The two *D. citri* parasitoids, *T. radiata* and *D. aligarhensis*, have preferences for different host stages: *T. radiata* lays eggs on 4th–5th stage nymphs, whereas *D. aligarhensis* lays eggs within the 2nd–4th stage nymphs (Milosavljević et al. 2017b). Thus, *T. radiata* and *D. aligarhensis*, may have potential to act in a complementary manner and collectively intensify biocontrol of *D. citri* in California through host resource, geographic, or habitat partitioning (Vankosky and Hoddle 2017a,b, Milosavljević et al. 2017b). The DD models developed here may greatly improve forecasting accuracy and the precision of inoculative (or augmentative) releases of mass reared parasitoids in response to expanding *D. citri* populations as they invade previously uninfested urban areas and commercial orchards such as the San Joaquin Valley in California. Moreover, should augmentative releases of *T. radiata* and *D. aligarhensis* become economically feasible in southern California, region-specific phenology models would assist with optimizing release timings aimed at maximizing the impact of released parasitoids.

Parasitism of large *D. citri* nymphs by *T. radiata* was found at all study sites. However, average percentage parasitism fluctuated substantially month to month across study sites. These observations are consistent with previous studies that show *T. radiata* parasitism rates vary greatly depending on location, time of year, and availability of appropriate life stages for parasitism (Tsai et al. 2002, Michaud 2004, Qureshi and Stansly 2009, Kistner et al. 2016a). Our analyses suggest that inoculative *T. radiata* releases into urban citrus should be concentrated between 350 and 2,400 DDs (i.e., mid March through April [290–430 DDs], from mid June through July [970–1,230 DDs], and during October [2,240–2,450 DDs]), when *D. citri* nymphs susceptible for host feeding and parasitism were most abundant during this study. During the course of this study no *D. aligarhensis* activity was observed across study sites. However, our results indicate that *D. aligarhensis* releases should be scheduled according to the temporal abundance of small *D. citri* nymphs susceptible for host feeding and parasitism, between 200 and 2,400 DDs. On a calendar date basis, the critical time intervals for inoculative *D. aligarhensis* releases that would maximize establishment likelihood in southern California were from February through March (200–320 DDs), during June (910–1,190 DDs), and from September to mid-October (2,210–2,410 DDs).

Somewhat surprisingly, population trends of *D. citri* adults and the subsequent population growth (i.e., numbers of oviposited eggs and densities of small nymphs) were not affected by *T. radiata* activity across seasons. One hypothesis is that *D. citri* adults are long lived and could have increasing densities on citrus hosts because of ongoing recruitment to the resident population of adults infesting study trees. A second explanatory hypothesis is compensatory survivorship of large nymphs, which results when fewer competing immature *D. citri* leads to increased numbers surviving (Dmitriew and Rowe 2007, Orizaola et al. 2014, Zhao et al. 2017). More research is needed to explore the potential importance of adult recruitment and/or compensatory survivorship on *D. citri* population growth and the effects this may have on pest population reduction by natural enemies in California.

At the present time, *D. citri*–CLas is predominantly an urban problem in California (Milosavljević et al. 2017b). Spillover of CLas-infected psyllids from residential infestation foci into commercial citrus areas, however, could exacerbate considerably the threat *D. citri*–CLas presents to the California citrus industry. In an attempt to help manage this possibility, we quantified the seasonal dynamics of key *D. citri* life stages to identify the most effective time to take action (e.g., inoculative or augmentative natural enemy releases, or spray applications) against pest populations in urban settings where *D. citri*–CLas complex primarily resides. The DD models, such as those used here, may have high utility for incipient area-wide management programs and the eventual development of IPM programs that aim to incorporate natural enemies with judicious pesticide use in commercial citrus.

Supplementary Material

Supplementary data are available at *Environmental Entomology* online.

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References Cited

Bayles, B. R., S. M. Thomas, G. S. Simmons, E. E. Grafton-Cardwell, and M. P. Daugherty. 2017. Spatiotemporal dynamics of the Southern California Asian citrus psyllid (*Diaphorina citri*) invasion. PLoS ONE. 12: e0173226.

Boina, D. R., W. L. Meyer, E. O. Onagbola, and L. L. Stelinski. 2009. Quantifying dispersal of *Diaphorina citri* (Hemiptera: Psyllidae) by immunomarking and potential impact of unmanaged groves on commercial citrus management. Environ. Entomol. 38: 1250–1258.

Bové, J. M. 2006. Huanglongbing: a destructive, newly-emerging, century-old disease of citrus. J. Plant Pathol. 88: 7–37.

Broatch, J. S., L. M. Dosdall, G. W. Clayton, K. N. Harker, and R. C. Yang. 2006. Using degree-day and logistic models to predict emergence patterns and seasonal flights of the cabbage maggot and seed corn maggot (Diptera: Anthomyiidae) in canola. Environ. Entomol. 35: 1166–1177.

Cammell, M. E., and J. D. Knight. 1992. Effects of climatic change on the population dynamics of crop pests. Adv. Ecol. Res 22: 117–162.

[CDFA] California Department of Food and Agriculture. 2017. CDFA ACP/HLB regulation and quarantine boundaries. https://www.cdfa.ca.gov/plant/acp/regulation.html.

[CIMIS] California Irrigation Management Information System. 2017. California Department of Water Resources' California Irrigation Management Information System. State of California, Sacramento, CA. http://www.cimis.water.ca.gov.

Clark, C., G. Edwards, and M. Friedlaender. 1973. Beverton-Holt model of a commercial fishery: optimal dynamics. J. Fish. Res. Board Can. 30: 1629–1640.

Dmitriew, C., and L. Rowe. 2007. Effects of early resource limitation and compensatory growth on lifetime fitness in the ladybird beetle (*Harmonia axyridis*). J. Evol. Biol. 20: 1298–1310.

Doerr, M. D., J. F. Brunner, and V. P. Jones. 2005. Phenology of *Lacanobia subjuncta* (Lepidoptera: Noctuidae) in Washington and Oregon apple orchards. J. Econ. Entomol. 98: 2100–2106.

Gottwald, T. R. 2010. Current epidemiological understanding of citrus huanglongbing. Annu. Rev. Phytopathol. 48: 119–139.

Grafton-Cardwell, E. E., L. L. Stelinski, and P. A. Stansly. 2013. Biology and management of Asian citrus psyllid, vector of the huanglongbing pathogens. Annu. Rev. Entomol. 58:413–432.

Halbert, S. E., and K. L. Manjunath. 2004. Asian citrus psyllids (Sternorrhyncha: Psyllidae) and greening disease of citrus: a literature review and assessment of risk in Florida. Fla. Entomol. 87: 330–353.

Hall, D. G., M. G. Hentz, and R. C. Adair Jr. 2008. Population ecology and phenology of *Diaphorina citri* (Hemiptera: Psyllidae) in two Florida citrus groves. Environ. Entomol. 37: 914–924.

Hall, D. G., E. J. Wenninger, and M. G. Hentz. 2011. Temperature studies with the Asian citrus psyllid, *Diaphorina citri* Kuwayama: cold hardiness and temperature thresholds for oviposition. J. Insect Sci. 11: 1–15.

Hall, D. G., M. L. Richardson, E. D. Ammar, and S. E. Halbert. 2013. Asian citrus psyllid, *Diaphorina citri*, vector of citrus huanglongbing disease. Entomol. Exp. Appl. 146: 207–223.

Hoddle, M. S. 2012. Foreign exploration for natural enemies of Asian citrus psyllid, *Diaphorina citri* (Hemiptera: Psyllidae), in the Punjab of Pakistan for use in a classical biological control program in California USA. Pak. Entomol. 34: 1–5.

Hoddle, M. S., and R. Pandey. 2014. Host range testing of *Tamarixia radiata* (Hymenoptera: Eulophidae) sourced from the Punjab of Pakistan for classical biological control of *Diaphorina citri* (Hemiptera: Liviidae: Euphyllurinae: Diaphorinini) in California. J. Econ. Entomol. 107: 125–136.

Hoddle, M. S., C. D. Hoddle, S. V. Triapitsyn, S. Z. Khan, and M. J. Arif. 2014. How many primary parasitoid species attack nymphs of *Diaphorina citri* (Hemiptera: Liviidae) in Punjab, Pakistan? Fla. Entomol. 97: 1825–1828.

Institute SAS. 2009. SAS/STAT, version 9.2 user's guide, 2nd ed. SAS Institute, Cary, NC.

Johnson, S. N., X. Zhang, J. W. Crawford, P. J. Gregory, and I. M. Young. 2007. Egg hatching and survival time of soil-dwelling insect larvae: a partial differential equation model and experimental validation. Ecol. Model. 202: 493–502.

Jones, V. P., and J. F. Brunner. 1993. Degree-day models. *In* Beers EH, Brunner JF, Willett MJ, Warner GM (eds.), Orchard pest management: a resource book for the Pacific Northwest. Good Fruit Grower, Yakima, WA.

Jones, V. P., C. C. Eastburn, T. D. Wilburn, and J. F. Brunner. 2005. Instar-specific phenology of *Pandemis pyrusana* and *Choristoneura rosaceana* (Lepidoptera: Tortricidae) in Washington apple orchards. J. Econ. Entomol. 98: 875–883.

Jones, V. P., T. R. Unruh, D. R. Horton, N. J. Mills, J. F. Brunner, E. H. Beers, and P. W. Shearer. 2009. Tree fruit IPM programs in the western United States: the challenge of enhancing biological control through intensive management. Pest. Manag. Sci. 65: 1305–1310.

Jones, V. P., J. F. Brunner, G. G. Grove, B. Petit, G. V. Tangren, and W. E. Jones. 2010. A web-based decision support system to enhance IPM programs in Washington tree fruit. Pest Manag. Sci. 66: 587–595.

Jones, V. P., R. Hilton, J. F. Brunner, W. J. Bentley, D. G. Alston, B. Barrett, R. A. Van Steenwyk, L. A. Hull, J. F. Walgenbach, W. W. Coates, and T. J. Smith. 2013. Predicting emergence of codling moth, *Cydia pomonella* (Lepidoptera: Tortricidae) in North America. Pest. Manag. Sci. 69: 1393–1398.

Joshi, N. K., E. G. Rajotte, K. J. Naithani, G. Krawczyk, and L. A. Hull. 2016. Population dynamics and flight phenology model of codling moth differ between commercial and abandoned apple orchard ecosystems. Front. Physiol. 7:408.

Khan, S. Z., M. J. Arif, C. D. Hoddle, and M. S. Hoddle. 2014. Phenology of Asian citrus psyllid (Hemiptera: Liviidae) and associated parasitoids on two species of citrus, kinnow mandarin and sweet orange, in Punjab Pakistan. Environ. Entomol. 43: 1145–1156.

Kistner, E. J., R. Amrich, M. Castillo, V. Strode, and M. S. Hoddle. 2016a. Phenology of Asian citrus psyllid (Hemiptera: Liviidae), with special reference to biological control by *Tamarixia radiata*, in the residential landscape of Southern California. J. Econ. Entomol. 109: 1047–1057.

Kistner, E. J., N. Melhem, E. Carpenter, M. Castillo, and M. S. Hoddle. 2016b. Abiotic and biotic mortality factors affecting Asian citrus psyllid (Hemiptera: Liviidae) demographics in Southern California. Ann. Entomol. Soc. Am. 109: 860–871.

Lewis-Rosenblum, H., X. Martini, S. Tiwari, and L. L. Stelinski. 2015. Seasonal movement patterns and long-range dispersal of Asian citrus psyllid in Florida Citrus. J. Econ. Entomol. 108: 3–10.

Liu, Y. H., and J. H. Tsai. 2000. Effects of temperature on biology and life table parameters of the Asian citrus psyllid, *Diaphorina citri* Kuwayama (Homoptera: Psyllidae). Ann. App. Biol. 137: 201–206.

Logan, J. D., W. Wolesensky, and A. Joern. 2006. Temperature-dependent phenology and predation in arthropod systems. Ecol. Model 196: 471–482.

Michaud, J. P. 2004. Natural mortality of Asian citrus psyllid (Homoptera: Psyllidae) in central Florida. Biol. Control 29: 260–269.

Milosavljević, I., A. D. Esser, and D. W. Crowder. 2016. Effects of environmental and agronomic factors on soil-dwelling pest communities in cereal crops. Agric. Ecosyst. Environ. 225: 192–198.

Milosavljević, I., A. D. Esser, and D. W. Crowder. 2017a. Seasonal population dynamics of wireworms in wheat crops in the Pacific Northwestern United States. J. Pest Sci. 90: 77–86.

Milosavljević, I., K. A. Schall, C. D. Hoddle, D. J. W. Morgan, and M. S. Hoddle. 2017b. Biocontrol program targets Asian citrus psyllid in California's urban areas. Calif. Agric. 71: 169–177.

Murtaugh, P. A., S. C. Emerson, P. B. McEvoy, and K. M. Higgs. 2012. The statistical analysis of insect phenology. Environ. Entomol. 41: 355–361.

Nava, D. E., M. L. G. Torres, M. D. L. Rodrigues, J. M. S. Bento, and J. R. P. Parra. 2007. Biology of *Diaphorina citri* (Hem., Psyllidae) on different hosts and at different temperatures. J. Appl. Entomol. 131: 709–715.

Nietschke, B. S., R. D. Magarey, D. M. Borchert, D. D. Calvin, and E. Jones. 2007. A developmental database to support insect phenology models. Crop. Prot. 26: 1444–1448.

O'hara, R. B., and D. J. Kotze. 2010. Do not log-transform count data. Methods Ecol. Evol. 1: 118–122.

Orizaola, G., E. Dahl, and A. Laurila. 2014. Compensatory growth strategies are affected by the strength of environmental time constraints in anuran larvae. Oecologia 174: 131–137.

Patt, J. M., and M. Setamou. 2010. Responses of the Asian citrus psyllid to volatiles emitted by the flushing shoots of its rutaceous host plants. Environ. Entomol. 39: 618–624.

Pedigo, L. P. 1999. Entomology and pest management, 3rd ed. Prentice Hall, Englewood Cliffs, NJ.

Qureshi, J. A., and P. A. Stansly. 2009. Exclusion techniques reveal significant biotic mortality suffered by Asian citrus psyllid *Diaphorina citri* (Hemiptera: Psyllidae) populations in Florida citrus. Biol. Control 50: 129–136.

Ricker, W. E. 1954. Stock and recruitment. J. Fish. Res. Board Can. 11: 559–623.

Robbins, P. S., R. T. Alessandro, L. L. Stelinski, and S. L. Lapointe. 2012. Volatile profiles of young leaves of Rutaceae spp. Varying in susceptibility to the Asian citrus psyllid (Hemiptera: Psyllidae). Fla. Entomol. 95: 774–776.

Ruppel, R. F. 1983. Cumulative insect-days as an index of crop protection. J. Econ. Entomol. 76: 375–377.

Sileshi, G. 2006. Selecting the right statistical model for analysis of insect count data by using information theoretic measures. Bull Entomol Res. 96: 479–488.

Thomas, S. M., G. S. Simmons, and M. P. Daugherty. 2017. Spatiotemporal distribution of an invasive insect in an urban landscape: introduction, establishment and impact. Landscape Ecol. 32: 2041–2057.

Tsai, J. H., and Y. H. Liu. 2000. Biology of *Diaphorina citri* (Homoptera: Psyllidae) on four host plants. J. Econ. Entomol. 93: 1721–1725.

Tsai, J. H., J. J. Wang, and Y. H. Liu. 2002. Seasonal abundance of the Asian citrus psyllid, *Diaphorina citri* (Homoptera: Psyllidae) in southern Florida. Fla. Entomol. 85: 446–451.

Udell, B. J., C. Monzo, T. M. Paris, S. A. Allan, and P. A. Stansly. 2017. Influence of limiting and regulating factors on populations of Asian citrus psyllid and the risk of insect and disease outbreaks. Ann. Appl. Biol. 171: 70–88.

Vankosky, M. A., and M. S. Hoddle. 2017a. An assessment of interspecific competition between two introduced parasitoids of *Diaphorina citri* (Hemiptera: Liviidae) on caged citrus plants. Insect Sci. doi:10.1111/1744–7917.12490.

Vankosky, M. A., and M. S. Hoddle. 2017b. The effects of conspecific and heterospecific interactions on foraging and oviposition behaviours of two parasitoids of *Diaphorina citri*. Biocontrol Sci Technol. 27: 739–754.

Ver Hoef, J. M., and P. L. Boveng. 2007. Quasi-Poisson vs. negative binomial regression: how should we model overdispersed count data? Ecology 88: 2766–2772.

Weibull, W. 1951. A statistical distribution function of wide applicability. J. Appl. Mech. 18: 293–297.

Whittingham, M. J., P. A. Stephens, R. B. Bradbury, and R. P. Freckleton. 2006. Why do we still use stepwise modelling in ecology and behaviour? J. Animal Ecol. 75: 1182–1189.

Yang, Y., M. Huang, C. G. A. Beattie, Y. Xia, G. Ouyang, and J. Xiong. 2006. Distribution, biology, ecology and control of the psyllid *Diaphorina citri* Kuwayama, a major pest of citrus: a status report for China. Int. J. Pest Manag. 52: 343–352.

Zhao, W., Z. You, J. Wei, and S. Wang. 2017. Compensatory population growth in *Daphniopsis tibetana* Sars (Crustacea: Cladocera) following starvation. Limnology 18: 167–174.