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Can parasitism by *Tamarixia radiata* (Hymenoptera: Eulophidae) affect the movement and oviposition behavior of *Diaphorina citri* (Hemiptera: Psyllidae)?

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Abstract *Diaphorina citri* is the vector of the bacterium that cause "huanglongbing" (HLB). The psyllid acquires the pathogen after feeding on infected plants and transmits it to other trees as it moves through the orchard. The psyllid's movement is directly affected by abiotic and biotic factors, such as the presence of conspecific insects and natural enemies. We evaluated the effects of the presence of conspecific nymphs either not parasitized or parasitized by *Tamarixia radiata* on the movement and oviposition of adult female *D. citri. Diaphorina citri* females were released into cages containing a row of four equidistant *Murraya paniculata* plants. The first plant, the release point, had one of three conditions: no nymphs; non-parasitized nymphs (high and low densities); or previously parasitized nymphs. At 24, 48, and 72 h following release, the females on each plant in the cage were counted. At the end of the observations (72 h), the females were removed, and the eggs laid on the plants were counted. The presence of non-parasitized nymphs, at low and high densities, increased the tendency of adult movement and reduced the total number of laid eggs, compared with cases in which nymphs were absent. On the other hand, the presence of nymphs increased the egg distribution by females over all available plants in the cages. In these cases, higher number of adult females and eggs were observed in plants without nymphs, compared with the plant in which nymphs were present (release point). The conditions of nymphs, i.e., if parasitized or not, did not change the movement behavior and the distribution of eggs deposited over plants in the cages. However, the number of eggs laid by females was low for the cases in which the nymphs were parasitized. The findings can contribute to establishing the optimal of application of the psyllid management techniques, and improving the efficiency of natural enemy releases considering that the pest can be present at the field in different life stages.

Keywords Aggregation · Intraspecific competition · Choice of oviposition and feeding site · Displacement · Asian citrus psyllid

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Introduction

Biotic and abiotic factors directly affect the ability of living beings to move, as they are decisive in determining the ability of species to disperse and survive (Dingle 1972; Stinner et al. 1983; Minter et al. 2018; Naranjo 2019). In this context, the movement of insect pests defines their spatial temporal abundance and distribution, and consequently the severity of the damage they cause to the attacked crops, as well the ecological and evolutionary impact on population regulation, community dynamics and gene flow (Mazzi and Dorn 2012). Understanding the effects of biotic and abiotic factors on the behavior of insects during their movement and search for oviposition and feeding sites can contribute to management programs for insect agricultural pests, such as the development of pest occurrence and prediction models or models that direct the best moment of application of the management tactics (Bullock et al. 2002; Mazzi and Dorn 2012; Garcia et al. 2019, 2021).

The Asian citrus psyllid *Diaphorina citri* transmits the bacterium that causes "huanglongbing" (HLB) is native to Asia. Due to its dispersal capacity, which increases from the immature to the adult stage (Sétamou et al. 2020), this species is now present on most continents (Halbert 1998; French et al. 2001; Bové 2006; Shimwela et al. 2016; Oke et al. 2020). Consider rewording...transport between continents is presumably human-aided

Although *D. citri* adults tend to remain in the same grove after their arrival, they continually move over short or long distances within the orchard (Boina et al. 2009; Kobori et al. 2011; Lewis-Rosenblum et al. 2015; Tomaseto et al. 2016, 2019). During these movements, adults and nymphs can acquire the bacterium (HLB) by feeding on an infected plant for 30 minutes or more (Capoor et al. 1974; Hung et al. 2004; Pelz-Stelinski et al. 2010). The adult becomes a HLB transmitter 3 days after feeding (Xu et al. 1988) and the nymphs transmit HLB after they become adults (Inoue et al. 2009; Pelz-Stelinski et al. 2010). Infected trees become unviable after 4–5 years, and their fruits drop before ripening and have lower sugar content (Grafton-Cardwell et al. 2013; Dala-Paula et al. 2019).

Considered the most destructive citrus disease, in 2014 HLB reduced orange production in Florida by approximately 43% (Singerman and Useche 2016) and has decimated more than 50 million trees in Brazil (CDA 2019). The disease significantly reduces citrus production in Africa (Garnier and Bové 1996; FAO 2002), Asia (Martinez and Wallace 1968; Bhavakul et al. 1982; Bové 1986; Sharma 2008; Shivankar et al. 2000), and Oceania (Fraser 1978). Infection reduction is achieved by eliminating infected plants, using healthy seedlings for planting, and managing the vector insect through application of chemical products and/or release of natural enemies, mainly *Tamarixia radiata* (Hymenoptera: Eulophidae), an ectoparasitoid that attacks the psyllid nymphs (Diniz et al. 2020; Garcia et al. 2021).

The movement of psyllids in grove is influenced by the presence and density of shoots available on the host, temperature, relative humidity, and wind (Yamamoto et al. 2001; Lewis-Rosenblum et al. 2015; Tomaseto et al. 2017; Garcia et al. 2019; Antolinez et al. 2021). In addition to these factors, the frequency and pattern of movement of *D. citri* adults in the search for host plants and the selection of microhabitats can also be influenced by competitive interactions with conspecifics and by encounters with natural enemies (Fritz 1982; Nufio and Papaj 2001). Natural control agents can, in addition to directly reducing populations, indirectly affect the behavior, physiology, morphology, and development of their prey/hosts. This indirect effect or non-consumptive effect was shiown in studies with parasites (Holmes and Bethel 1972; Price 1980) and predators that found reductions in the population growth of aphids, *Acyrthosiphon pisum* (Homoptera: Aphididae) by damsel bugs (Hemiptera: Nabidae) (Nelson et al. 2004), *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae) by *Podisus maculiventris* (Heteroptera: Pentatomidae) (Hermann and Thaler 2014), and *Diaphorina citri* by *Hippodamia convergens* (Coleoptera: Coccinelidae) (Seo et al. 2018).

Since most of the *D. citri* primary inoculum sites are external to commercial orchards (Tiwari et al. 2010), knowledge of parameters that will stimulate the start of psyllid migration from these sites as well as of the effects of intraspecific competition (i.e., density of conspecifics) and the presence of natural enemies on the pattern of movement and oviposition of *D. citri* adults can contribute to the improvement of techniques used in integrated management (IPM) of this insect pest, for example, to define pest sampling techniques, as well the distribution of release points for natural enemy releases in the field. This study evaluated the movement of psyllid females and the pattern of egg distribution in experimental cages containing host plants in different situations: (*i*) healthy plants (*i.e.,* without psyllid nymphs), (*ii*) plants with *D. citri* nymphs, and *(iii*) plants with *D. citri* nymphs parasitized by *T. radiata*.

Materials and methods

Colony of insects and host plants

For this study, nymphs and adults (females) of *D. citri* and adults of *T. radiata* were obtained from the rearing colonies at the Laboratory of Insect Biology of the University of São Paulo –ESALQ/USP, Piracicaba, maintained following the method of Parra et al. (2016). The host plants used were orange jessamine, *Murraya paniculata* (L.) Jack, kept in a greenhouse under natural conditions and with automatic ventilation that maintains the temperature between 25 and 28 °C.

Experimental design

To evaluate the effects of the presence of conspecifics and the presence of parasitized nymphs on the movement and oviposition pattern of *D. citri* during the search for available plants for food and development of its offspring, a group of 90 adult psyllid females, 9-days old (average period of onset of oviposition) (Nava et al. 2007), was used. Prior to the beginning of the experiment, females were kept for 9 days in cages containing males, to ensure mating. After this period, the 90 females were selected randomly from the cages and but you just mentioned that there were (only) 90 females in the cagetransferred to fabric tunnel cages (200 × 60 × 70 cm), each containing 4 tubes (250 ml) with 4 orange jessamine shoots per tube and spaced equidistantly from each other (Fig. 1). The shoots had an average length of 1 cm, as this is the most attractive size for oviposition (Diniz 2013). After the insects were released, the number of females on each plant (A, B, C, and D) was counted to assess the movement of insects on the available plants over 24, 48, and 72 h. At the end of 72 h, the total number of eggs laid on each plant was also counted. The cages were kept in the greenhouse under the same conditions described above.

In the first set of experiments, the effect of intraspecific competition on the movement and distribution of *D. citri* eggs was evaluated. Inside the experimental cages, 4th and 5th instar psyllid nymphs were placed on the first plant (plant A, release site). Two nymph densities (*i.e.,* treatments) were established*: (i)* low density, with 15 nymphs present; and *(ii)* high density, with 70 nymphs present. A control consisted of plants with no nymphs. Ten replicates were performed for each treatment and control.

In the second set of experiments, the effect of the presence of parasitized nymphs on the movement behavior of *D. citri* and its eggs distribution was evaluated, using two treatments: *(i)* presence of 25 non-parasitized *D. citri* nymphs on plant A and *(ii)* presence of 25 nymphs previously parasitized by *T. radiata* on plant A. Ten replicates were performed for each treatment. Treatment without the nymphs was done in the first experiment. The parasitized nymphs were easily recognized by the mummified aspect, by the presence of meconium and wax around the body (Fig. 4).

Data analyses

For the analysis of the movement of insects between plants over time (24, 48, and 72 h) an extension of the discrete-time transition models for clustered nominal data was used, according to the procedure proposed by Lara et al. (2020). In this procedure, a multinomial

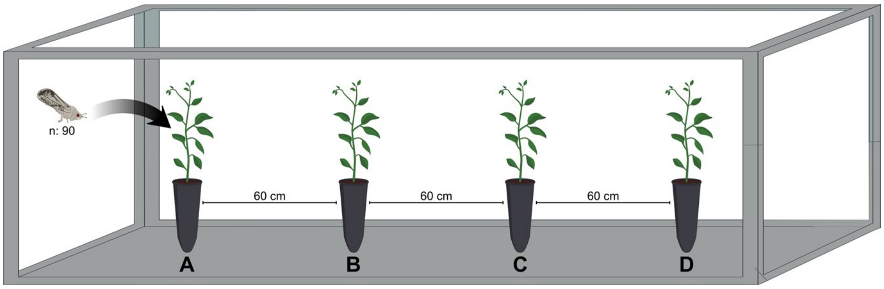

Fig. 1 Experimental cage to evaluate the movement and distribution pattern of *Diaphorina citri* eggs on *Murraya paniculata* plants

distribution was assumed for the response variable, with five categories: Plant A, Plant B, Plant C, Plant D, and none (insects that were in the cage but not on one of the plants), each representing the preference of insect groups, and the generalized logit models were fitted (Agresti 2013). The first-order Markov stochastic assumption was used to fit the model, i.e., the number of insects on each of the plants was incorporated into the linear predictor of the model as additional covariates. To decide between a stationary process or not, the likelihood ratio test proposed by Lara et al. (2017) was used. If the stationary model is assumed, in practice, insects move through the vases, following a homogeneous pattern over time.

For the analysis of the variation of the total number of eggs deposited on the four plants (A, B, C, and D) after 72 h, considering the possible effect of treatments (experiment 1: high nymph densities, low nymph density, and absence of nymphs; experiment 2: parasitized nymphs and non-parasitized nymphs), the Generalized Linear Model was fitted assuming a Poisson distribution, considering the Quasi-Poisson model when over dispersion is found, which is a common phenomenon with count data with a variance much greater than the mean (McCullagh and Nelder 1989). The best statistical model was chosen through comparisons between the adjustments of the complete models and models with the reduction of explanatory variables by the Akaike criterion (AIC). The quality of the adjustments obtained by the models was assessed using the “half-normal-plot” graph with simulated envelopes at the 95% level (Moral et al. 2017). All analyses were performed using the R software (R Core Team 2019).

Results

Experiment 1: Movement rate of females and egg distribution pattern of *Diaphorina citri* in the presence of non-parasitized nymphs

Time variation (24, 48, and 72 h), covariates for psyllid occupation options (plants A, B, C, D, and none), and the presence of nymphs (low or high density) significantly affected the movement of the adult females of *D. citri* (LRV = 55.34, df = 28, $p < 0.01$; LRV = 746.67, df = 16, $p < 0.01$; and LRV = 13640.51, df = 8, $p < 0.01$, respectively). The results showed that in the control group, in which nymphs were absent, the average proportion of insects remained higher on plant A (insect release point) during the three evaluation periods (24, 48, and 72 h) compared to the proportion of insects that remained on plant A when nymphs were present (Fig. 2, Table 1). At low and high nymph densities with the passage of time, females tended to move toward the other plants in the cages (Table 1). The proportion of psyllids elsewhere than on the plants was

Fig. 2 Mean percentages of adult females of *Diaphorina citri* on *Murraya paniculata* plants in the absence and presence of nymphs in low and high densities over 24, 48, and 72 h

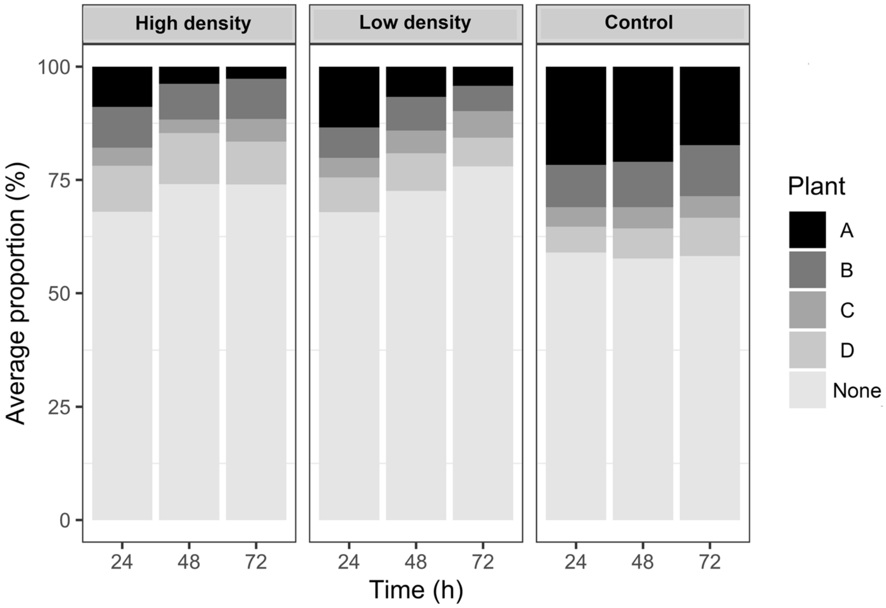

Table 1 Numbers (mean ± standard error) of insects found on the four plants (A, B, C, D) after release of *Diaphorina citri* on plant A (with the presence or absence of nymphs in low or high density), over 3 days

Treatment	Period (h)	Mean no. of insects			
		Plant A	Plant B	Plant C	Plant D
Absence of nymphs (Control)	24	19.5 ± (2.5) Aa	8.4 ± (1.9) Aab	3.9 ± (1.2) Ab	5.1 ± (1.1) Ab
	48	18.9 ± (2.3) Aa	9.0 ± (2.3) Aab	4.2 ± (0.9) Ab	6.0 ± (1.3) Ab
	72	15.6 ± (2.3) Aa	10.1 ± (2.1) Aab	4.3 ± (1.1) Ab	7.6 ± (1.7) Ab
Low nymph density	24	12.1 ± (2.8) Aa	6.0 ± (1.1) Aab	3.9 ± (1.1) Ab	6.9 ± (2.0) Aab
	48	6.0 ± (2.1) ABa	6.7 ± (0.9) Aa	4.5 ± (1.3) Aa	7.5 ± (2.1) Aa
	72	3.8 ± (1.6) Ba	5.0 ± (1.1) Aa	5.3 ± (1.5) Aa	5.7 ± (1.9) Aa
High nymph density	24	8.0 ± (1.6) Aa	8.1 ± (1.8) Aa	3.6 ± (0.6) Ab	9.1 ± (1.6) Aa
	48	3.4 ± (0.9) Bb	7.1 ± (2.0) Aab	2.7 ± (0.5) Ab	10.1 ± (2.0) Aa
	72	2.4 ± (1.1) Bb	8.0 ± (2.1) Aa	4.5 ± (0.7) Aab	8.5 ± (2.1) Aa

Means followed by different letters indicate a significant difference at significance level α = 5% (likelihood ratio test). Uppercase letter compares the times inside plants for each treatment (rows), lowercase letter compares the plants at each time for each treatment (column)

also higher and increased with time when nymphs were present, indicating a greater tendency to move compared to the control group (Fig. 2).

The presence of nymphs (low or high density) and the distance of the plants from the release point (plant A) significantly affected the distribution of eggs on the different plants ($F_{6,108}$= 3.83, $p < 0.01$). In the control group, the highest number of eggs was found on plant A. In plants B, C, and D, fewer eggs were laid and the numbers of eggs deposited on these plants did not differ significantly (Table 2). The presence of nymphs at the release point (plant A) resulted in changes in the distribution of eggs by females. When the density of nymphs on plant A was low, the distribution of eggs was more uniform among the plants in the cage, with no significant difference among plants. However, when the density of nymphs on plant A was high, more eggs were observed on plants D and B, in that order, 72 h after the females were released into the cages (Table 2).

Experiment 2: Movement rate of females and pattern of egg distribution of *Diaphorina citri* in the presence of non-parasitized nymphs and nymphs parasitized by *Tamarixia radiata*

The variation in time (24, 48, and 72 h), the covariates for occupation options by the psyllid (plants A, B, C, D, and none), and the presence of nymphs (parasitized or not parasitized) significantly affected the movement on the plants by adult females of *D. citri* (LRV = 60.44, df = 24, $p < 0.01$; LRV = 405.92, df = 16, $p < 0.01$, and LRV = 3258.7, df = 4, $p < 0.01$, respectively). The results showed that number of adult psyllids on each plant in the cage did not differ over the observation period (24, 48 and 72h) (Fig. 3 and Table 3). However, comparing the proportion of psyllids among the plants on each period of time, when parasitized nymphs were present, the adult psyllids showed the tendency to remain in plant A. On the other hand, when nymphs were not parasitized, the insects tendend to move toward the other plants in the cages (Fig. 3, Table 3).

Regarding the distribution of eggs on the four plants, there was no effect of plant and parasitism interaction ($F_{3,72}$ = 0.3017, p = 0.8241). The plant effect was also not significant ($F_{3,75}$= 0.0349, p = 0.9912). However, the presence of non-parasitized or parasitized nymphs had a non-significant but marginal effect ($F_{1,78}$ = 3.74, p = 0.0571) on the distribution of eggs on the four plants. The mean number of eggs was high in cages in which nymphs were not parasitized (Table 4A). There was no significant difference between the number of eggs deposited among plants (Table 4B). When the present nymphs on plant A were parasitized, females laid their eggs under nymphs (Fig. 4), differing from the other cases, in which the eggs were laid on the plant shoots.

Table 2 Numbers (mean ± SD) of eggs of *Diaphorina citri* found on the four plants (A, B, C, D), in the presence (low or high density) or absence of nymphs on plant A (insect release point) 72 h after adults were released

Treatment	Mean no. of eggs				Total
	Plant A	Plant B	Plant C	Plant D	
Absence of nymphs (Control)	205.7 (± 63.5) Aa	74.7 (± 22.7) Ba	42.1 (± 13.7) Ba	58.2 (± 31.3) Ba	3807 A
Low density of nymphs	26.6 (± 10.3) Ab	49.6 (± 16.4) Aa	27.4 (± 10.9) Aa	39.8 (± 20.5) Aa	1434 B
High density of nymphs	8.9 (± 3.9) Ab	87.0 (± 41.7) Ba	27.1 (± 12.4) Aa	93.9 (± 30.8) Ba	2169 B
Total	2412	2113	966	1919	

Means followed by different letters indicate a significant difference in the number of eggs at significance level α = 5% (likelihood ratio test). Lowercase letter compares each plant among the treatments (row) and uppercase letter compares the different plants within each treatment (column). For total marginal, uppercase letter compares treatments, regardless of plant (row)

Fig. 3 Mean percentages of adult females of *Diaphorina citri* on *Murraya paniculata* plants in the presence of non-parasitized or parasitized nymphs over 24, 48, and 72 h

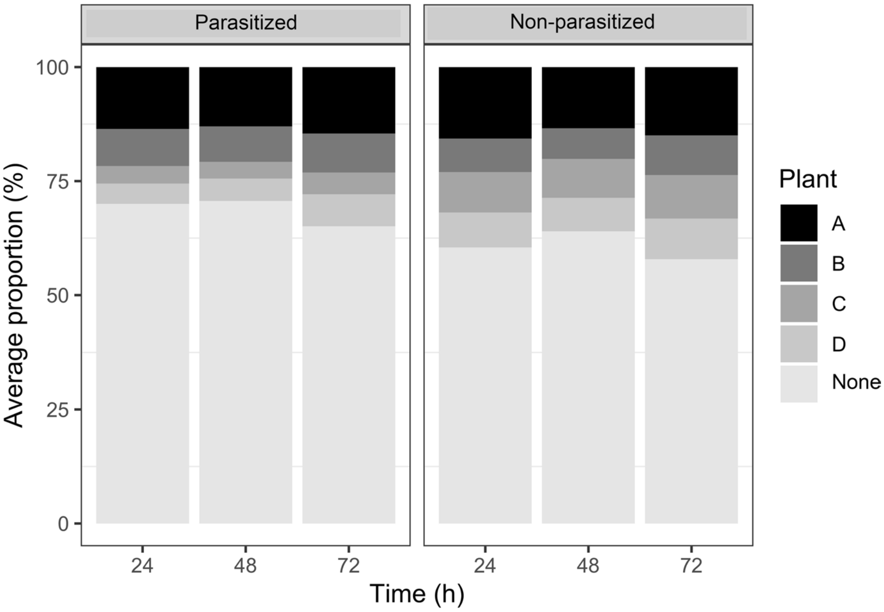

Table 3 Numbers (mean ± standard error) of insects, *Diaphorina citri*, found on the four plants (A, B, C, D), in the presence of non-parasitized or parasitized nymphs on plant A, over 3 days

Treatment	Period (h)	Mean no. of insects			
		Plant A	Plant B	Plant C	Plant D
Non-parasitized nymphs	24	14.1 ± 2.7 Aa	6.6 ± 1.1 Ab	8.0 ± 1.6 Aab	6.9 ± 1.0 Ab
	48	12.1 ± 2.2 Aa	6.0 ± 1.2 Ab	7.7 ± 1.4 Aab	6.6 ± 0.7 Ab
	72	13.5 ± 2.5 Aa	7.8 ± 1.7 Aa	8.6 ± 1.8 Aa	8.0 ± 1.3 Aa
Parasitized nymphs	24	12.2 ± 1.5 Aa	7.3 ± 1.5 Ab	3.5 ± 0.7 Ac	4.0 ± 0.5 Ac
	48	11.7 ± 1.5 Aa	7.0 ± 1.4 Ab	3.3 ± 0.7 Ac	4.4 ± 0.5 Ac
	72	13.1 ± 1.9 Aa	7.7 ± 1.6 Aab	4.3 ± 1.1 Ab	6.3 ± 1.2 Ab

Means followed by different letters indicate a significant difference at significance level α = 5% (likelihood ratio test). Uppercase letter compares the times inside plants for each treatment (rows), lowercase letter compares the plants at each time for each treatment (column)

Table 4 Numbers (mean ± SD) of eggs of *Diaphorina citri* found on the four plants (A, B, C, D), in the presence of non-parasitized or parasitized nymphs on plant A, 72 h after the release of adults

(A) Treatment	Mean	(B) Mean nº of eggs			
		Plant A	Plant B	Plant C	Plant D
Non-parasitized nymphs	49.9 (± 7.7) a	40.5 (± 11.2) A	41.9 (± 12.4) A	37.6 (± 10.7) A	40.0 (± 6.9) A
Parasitized nymphs	30.0 (± 6.5) b				
Total		810	839	747	800

Means followed by different letters indicate a significant difference at the significance level α = 5% (likelihood ratio test). Lowercase letter compares the treatments (row) and uppercase letter compares the different plants (column)

Discussion

In this study, the presence of *D. citri* nymphs affected the movement behavior of adults. In the absence of nymphs, adult females tended to remain at the release site (plant A). When nymphs (parasitized or non-parasitized) were present at the release site (plant A), the adult females tended to disperse to other plants, reducing aggregation. The presence of nymphs also altered the females' oviposition behavior. In the absence of nymphs, oviposition rates were higher on the plant with the highest aggregation of adults (plant A). However, when non-parasitized or parasitized nymphs were present on plant A, the number of eggs deposited on plant A was reduced.

Psyllid females showed aggregative behavior at the release site (plant A) in situations where nymphs were absent or present in low/medium densities. The tendency to remain at the place of release or arrival in an orchard was also observed in the field by Arakawa and Miyamoto (2007) in wind-tunnel experiments, and by Boina et al. (2009), who used protein markers to observe the bidirectional movement of insects between managed and unmanaged areas. These results support the idea that psyllids have low active mobility, even though they have a flight capacity of up to 2.4 km (Martini et al. 2014a).

The aggregative behavior observed may also be due to the attraction of *D. citri* to volatiles emitted by plants damaged by feeding of conspecifics present (Groot et al. 2003; Blackmer et al. 2004). In addition to the psyllid's attraction to citrus plants and orange jasmine (Patt and Sétamou 2010), its attraction to psyllid-damaged plants and its aggregation behavior were observed by Hijaz (2013) and Patt et al. (2018), who linked this result to increased release of salicylic acid, as previously found by Mann et al. (2012) with citrus. Although the study was carried out with plants infected with HLB, they observed an increase in methyl salicylate emission in plants that attracted more insects. Martini et al. (2014b), in their study with curry plants, also observed aggregated behavior of females of *D. citri* on damaged plants when fewer than 10 individuals

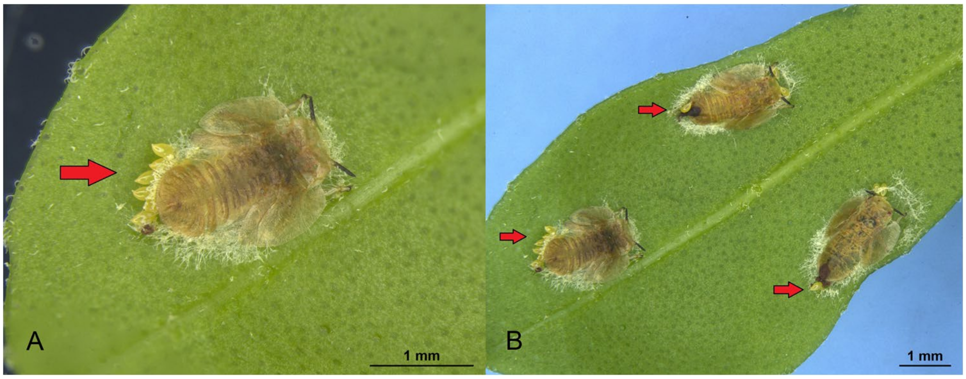

Fig. 4 **A** and **B** *Diaphorina citri* eggs under psyllid nymphs parasitized by *Tamarixia radiata*. Parasitism is evidenced by the presence of meconium and wax, on the back and around the body of the nymph, respectively

(females) were present. In the absence of damaged plants, the presence of more than 10 females had a repellent effect on other females, which were also not attracted by the presence of males of the species, although males were attracted by females.

In the absence of nymphs at the release site, the present results indicate that plants with more psyllids also had more eggs deposited. In this species, as in others such as *Cassida rubiginosa* (Coleoptera: Chrysomelidae) (Fujiyama et al. 2008) and *Thaumastocoris peregrinus* (Hemiptera: Thaumastocoridae) (Martínez and Bianchi 2010; Wilcken et al. 2010), the feeding and oviposition sites overlap, since *D. citri* adults and nymphs both prefer young growing shoots (Hall et al. 2013; Cifuentes-Arenas et al. 2018). This aggregation behavior of the adult and nymphal stages benefits the species, since it indicates better local abiotic conditions for development and reproduction (Sétamou et al. 2016; Cifuentes-Arenas et al. 2018). On the other hand, this aggregation also entails competition for space and food, in addition to increasing the risk of infections and the action of natural enemies (Martini et al. 2014b), which may explain the change in the adults' behavior as the process continued, with an increase in the density of nymphs on plant A.

Knowing that the presence of conspecifics at a location indicates quality and the degree of competition, a decrease in numbers of both adult and immature insects is expected at these locations (Davis et al. 2011), as was observed here in the treatments with low and high density of nymphs. Bawin et al. (2014), Guo et al. (2014), and Kohandani et al. (2015) found similar results and determined that the presence of conspecifics above a certain threshold is required to trigger a "displacer" effect on insects. Although this "displacer" effect was observed in this study in both experimental scenarios (presence of non-parasitized nymphs and presence of parasitized nymphs; Tables 1 and 2), eggs were still deposited at these "occupied" sites, even if other plants of the same quality were available for feeding and oviposition in the same cage.

Studies on the impact of the presence of natural enemies on the reproductive rate of hosts/prey have shown that *D. citri* can reduce the number of offspring due to the presence of a control agent (Holmes and Bethel 1972; Price 1980; Nelson et al. 2004; Ninkovic et al. 2013; Hermann and Thaler 2014). In this study the number of eggs in a presence of parasitized nymphs was lower than the number observed in cages with non-parasitized nymphs, although the analysis did not detect the effect of parasitism on number of laid eggs (p-value was marginally non-significant). However, in the presence of parasitized nymphs, *D. citri* laid eggs in the presence of the parasitoid, although the parasitoid was still developing within the psyllid nymphs (Fauvergue and Quilici 1991; Hoy and Nguyen 2000). *Diaphorina citri* also showed this behavior in another study (Seo et al. 2018), where it perceived and was repelled by the chemical traces of a natural enemy, the predator *H. convergens*, reducing its oviposition rate and excretions.

Detection of the presence of natural enemies has important consequences for the survival rate of an insect and can manifest at the level of predator/prey and host/parasitoid interactions as well as in intraguild competition (predator/parasitoid) interactions. For example, Milosavljević et al. (2021) found that the presence of *Linepithema humile*(Hymenoptera: Formicidae) strongly reduced the control of *D. citri* by *T. radiata*, since the ants defended the psyllids from the parasitoids in exchange for food. Thus, the development of behaviors by insects that minimize their risks in the presence of conspecifics and natural enemies is necessary from a biological point of view. For example, *T. radiata* marks the nymphs of *D. citri* with a pheromone after parasitizing them, to prevent superparasistism (Mann et al. 2010) and can detect volatiles to avoid predating of their nymphs (Milosavljević et al. 2021).

The results obtained here reinforce the recommendation that *D. citri* control tactics should be focused on the nymphal stages, since in addition to contributing more effectively to the spread of HLB (Inoue et al. 2009, Diniz et al. 2021), the presence of nymphs can stimulate adults to disperse, also contributing to the spread of the disease.

Conclusion

The results showed that *D. citri* has an aggregated behavior, with a tendency to low mobility in the absence of nymphs. However, this behavior changes Explain? when nymphs are present, in low or high densities and whether or not the nymphs are parasitized. *D. citri* distinguishes unparasitized nymphs from nymphs parasitized by *T. radiata*, and in this case, lays eggs close to the parasitized nymphs.

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Declarations

Conflict of interest The authors have no conflict of interest to declare.

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