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Quantification of direct and indirect damage caused by *Diceroprocta bulgara* (Distant) (Hemiptera: Cicadidae) in lime

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ABSTRACT

Objective: To quantify the direct and indirect damage caused by *Diceroprocta bulgara* in lime cultivars: Persian, Mexican with and without thorns, Colimex and Lise; on the Coast of Oaxaca, Mexico.

Design/Methodology/Approach: The damage caused by *D. bulgara* was calculated by counting nests in trees of five lime cultivars (Persian, Mexican with thorns, Mexican without thorns, Colimex and Lise). In each plot, 15 trees were selected randomly to evaluate the variables: height, crown diameter, damaged growth flows, damaged growth diameter, damage length, and number of eggs. The loss of fruit from the indirect effect (kg ha^{-1}), was estimated in three densities of Colimex and Lise.

Results: The damage ranged between 78.6% (Mexican without thorns) and 94% (Colimex). The trees of largest size showed a higher percentage of affected flows. The second flow showed the highest frequency of damage (32%), while the lowest frequency was obtained in the first flow (17.3%). The lowest number of eggs was 26 in flow one in Mexican lime without thorns, while the highest was 171 in Mexican lime without thorns. The greatest number of eggs was observed in Persian lime (371). A significant relationship was determined between the number of eggs and the damage length and flow diameter (<0.05). The highest number of mummified fruits (15.6) was observed in Colimex (density of $312 \text{ trees ha}^{-1}$) and the estimation that up to 146 kg of fruit could be lost.

Findings/Conclusions: This study quantifies for the first time the direct and indirect damage caused by oviposition of the cicada *D. bulgara*, which represents a loss of fruit in productive trees. Studies on the insect's biology are suggested for integral management plans.

Keywords: Citruses, fruit loss, cicada.

INTRODUCTION

Mexico was the main producer of lime in the world during the production cycle 2018-2019, with a production volume of 2.6 million tons, corresponding to 34% of global production (USDA/FAS, 2020). Two species of lime are cultivated in Mexico: *Citrus aurantifolia* (Christm.)



Swingle, and *Citrus latifolia* Tanaka ex Q. Jiménez; the first is distributed primarily in the Pacific watershed and the second in the Gulf of Mexico (Orozco-Santos *et al.*, 2013). In Oaxaca, Mexico, the Central Valleys Experimental Field, Coast of Oaxaca Experimental Site of the National Institute for Forestry, Agriculture and Livestock Research (*Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias*, INIFAP), has evaluated varieties of lime that adapt to the agroecological conditions of the Coast region, which allow diversifying the alternatives for production and commercialization in international markets (Ovando-Cruz *et al.*, 2015).

The main insect pests reported for the lime crop in Mexico are: psyllid (*Diaphorina citri* Kuwayama), leaf miner (*Phyllocnistis citrella* Stainton), white fly (*Dialeurodes* sp.), blackfly (*Aleurocanthus woglumi* (Ashby)) and some species of aphids (*Aphis spiraeicola* Patch and *Toxoptera aurantii* Boyer) (Grafton-Cardwell *et al.*, 2013; Ovando-Cruz *et al.*, 2015).

Orozco-Santos *et al.* (2014) reported the presence and possible damage associated to a cicada (Hemiptera: Cicadidae) in lime plants in Colima, México. Then, Sánchez-García *et al.* (2019) identified this species as *Diceroprocta bulgara* (Distant) and reported various citrus as hosts for oviposition. During this process, females cause longitudinal lesions on the bark of the trees' apical branches, which later leads to their death.

This study quantified the direct and indirect damage caused by oviposition of *D. bulgara* in cultivars of Persian lime and Mexican lime with and without thorns, as well as in the INIFAP varieties Colimex and Lise; the study also discusses the implications of damage caused by the insect and possible alternatives for control.

MATERIALS AND METHODS

Study area

The study was conducted in two plots with lime of INIFAP's Coast of Oaxaca Experimental Site, located in Río Grande, Villa de Tututepec, Oaxaca, Mexico. The first plot with one-year-old plants of the cultivars: Mexican with and without thorns, and Persian. The plants were established under a design with strips with distances of 4 m between plants and 8 m between lines. The second plot with three-year-old plants of the varieties Colimex and Lise, established in a system of strips with distances of 8×4 m, 7×4 m and 6×4 m.

Evaluation of the damage caused by cicadas

The evaluation was performed in plants from the two plots selected. In the first place, the presence of damage from *D. bulgara* (nests) was found in each tree of the five citrus cultivars. Then, 15 trees were selected randomly in each plot, of which data were taken of tree height and crown diameter; the number of flows with damaged vegetative growth from the cicada and the number of terminal branches affected in each flow were quantified. The diameter of the vegetative flow and the length of the damage were also measured in each of the samples, and the eggs laid were counted. The presence of at least one oviposition zone in one of the last three growth flows was considered as a characteristic to consider a damaged flow. The severity of the attack from cicadas was classified according to the number of flows with oviposition, and eight damage scales were proposed for that purpose:

Class 1=without damage, Class 2=1 to 5 flows, Class 3=6 to 10, Class 4=11 to 15, Class 5=16 to 20, Class 6=21 to 25, Class 7=26 to 30, and Class 8=more than 30.

The number of fruits affected indirectly by *D. bulgara* was recorded in the plants in productive stage, and an estimation of loss of fruit was conducted (kg ha^{-1}) based on the plantation density.

Statistical analysis

The variables stem diameter, damage length and number of eggs were analyzed through a multiple linear regression model with the software SAS version 10.0. This analysis was confirmed with tolerance (T) higher than 0.10, as variance inflation factor (VIF) lower than 5%, and the significant value of F and validity of the model by the lowest value of the Akaike information criterion (AIC).

RESULTS AND DISCUSSION

Oviposition by *D. bulgara* was observed in the two experimental plots. The process of oviposition generated partial damage in the vascular bundles of the affected shoots, which led to the death of the vegetative flow. This process was previously reported by Sánchez-García *et al.* (2019). The period of oviposition coincided with the phenological phases of flowering and fructification of the plots, which indirectly caused the death of small flowers and fruits close to harvest (Figures 1 and 2).

Table 1 presents the percentage of incidence of citrus trees affected by *D. bulgara*. The incidence of damage was quantified from 78.6 to 94%, based on the number of trees from each cultivar.

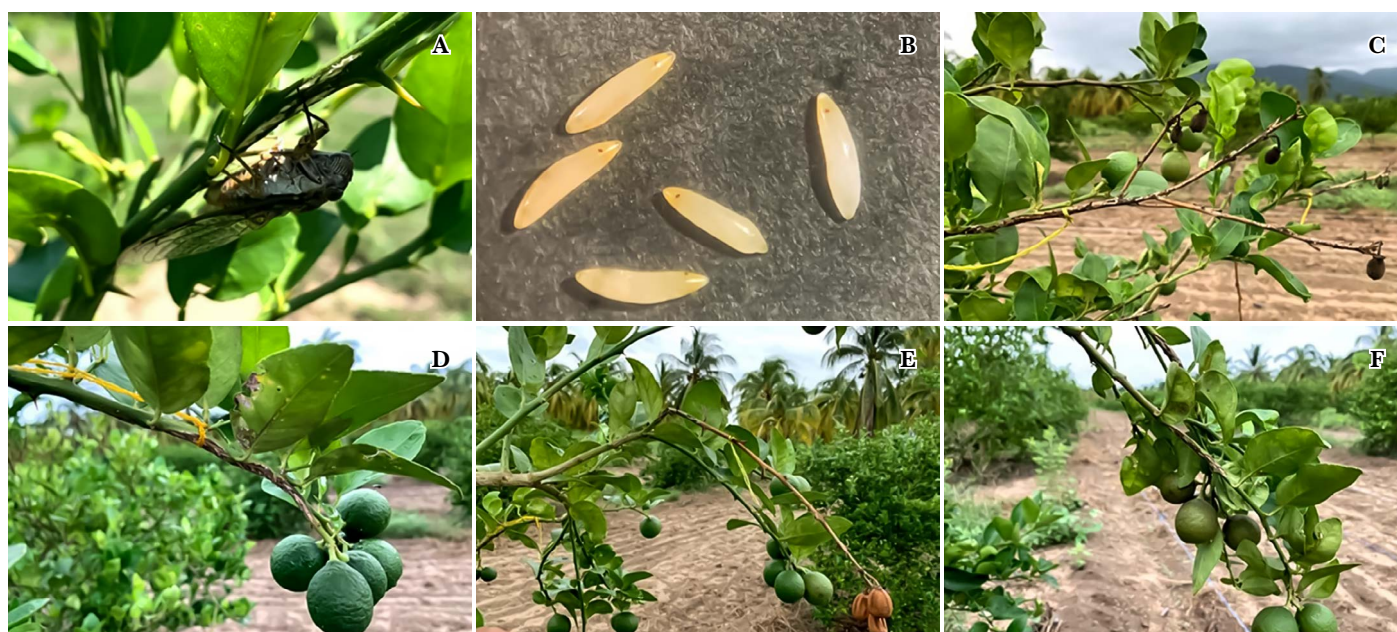


Figure 1. Female cicada of *D. bulgara* in Mexican lime plant (A), eggs (B) and damage caused in terminal flows and lime fruits as result of the oviposition of eggs (C-F). Río Grande, Villa de Tututepec, Oaxaca, June 2020.

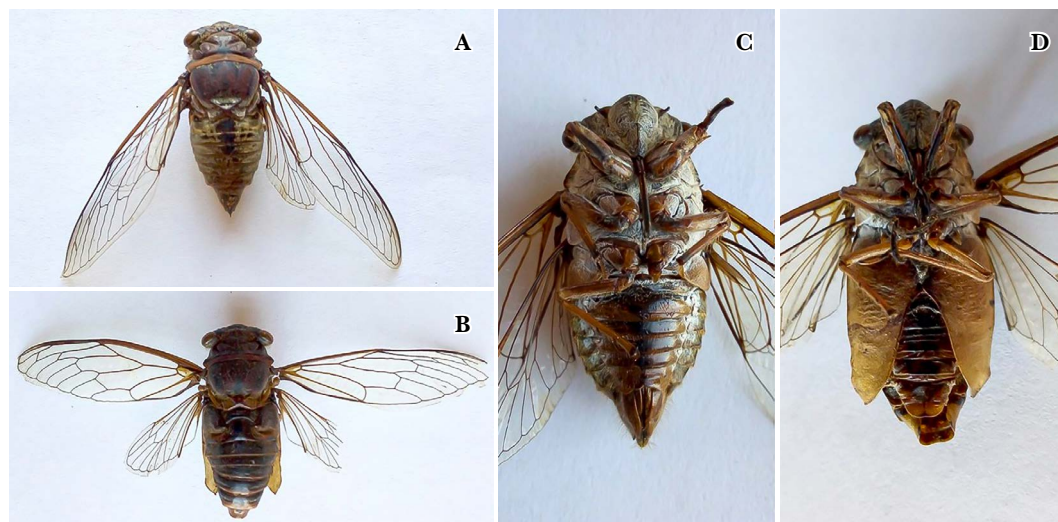


Figure 2. General aspect of the male and the female of *D. bulgara*. Female (A and C); male (B and D).

Table 1. Incidence of the damage by *D. bulgara* in trees from five citrus cultivars.

Cultivar	Number of tres (n)	Trees with damage (%)
Persa lime	178	83.70
Mexican with thorns	224	83.03
Mexican without thorns	224	78.57
Colimex	320	94.06
Lise	286	90.21

Regarding the relationship between the size and diameter of the trees with the average of damaged flows, a low correlation was observed where larger plants showed a higher percentage of affected shoots (Table 2). This can be attributed to the cicada females having a greater area or leaf cover to carry out oviposition compared to smaller plants. On the other hand, the average of affected terminal branches fluctuated negatively in terms of size of the trees (Table 2).

Table 2. Height and crown diameter and terminal growth flows (1-3) of lime trees with damage by *D. bulgara*.

Cultivar	Plant height (cm)	Crown diameter (cm)	Average number of damaged flows	Average number of affected terminal branches
Persa lime	122.40	72.73	2.33	4.73
Mexican with thorns	124.26	94.50	2.35	4.11
Mexican without thorns	137.00	70.83	2.06	3.40
Colimex	171.68	201.83	4.30	3.73
Lise	192.57	255.00	3.40	3.56

In the five citrus cultivars evaluated, damage by the cicada was observed in the three terminal flows. The flow with highest damage frequency was flow 2 (average of 32%) and the one with lowest frequency was flow 1 (average of 17.3%) (Table 3).

The average number of eggs laid per each flow evaluated was variable for each citrus variety, with a minimum of 26 for flow 1 in the cultivar Mexican without thorns, and a maximum of 171 in the same cultivar (Table 4). In general, a higher number of eggs was counted in Persian lime (371 eggs), compared to the Mexican lime with thorns (245 eggs). These values are higher than those reported by Sánchez-García *et al.* (2019), who counted an average of 48.4 eggs in Mexican lime.

The criteria used in the statistical analysis by multiple linear regression were accepted, based on the assumption of non-collinearity between the variables, through the criteria of tolerance values ($T > 0.10$) and the variance inflation factor ($VIF < 5.0$) (Table 5). Likewise, in the analysis of variance, the statistic F was highly significant (< 0.05), which indicates that there was significant linear relationship of the dependent variable number of eggs and the variables damage length and flow diameter.

Table 3. Frequency of oviposition by *D. bulgara* in the last three growth flows in five lime cultivars.

Cultivar	Samples (number)	Flow 1	Flow 2	Flow 3
Persa lime	15	15.55	28.88	20.00
Mexican with thorns	15	20.00	33.33	33.33
Mexican without thorns	15	17.77	33.33	33.33
Colimex	15	20.00	31.11	33.33
Lise	15	13.33	33.33	31.11

Table 4. Damage length, flow diameter and eggs per growth flow in five lime cultivars.

Cultivar	Flow 1			Flow 2			Flow 3			TH
	L	D	H	L	D	H	L	D	H	
Persa lime	10.0	1.9	82.8	11.4	2.8	128.4	10.6	4.4	160.6	371.8
Mexican with thorns	3.3	1.5	26.7	8.8	2.5	143.0	8.1	3.9	171.0	340.7
Mexican without thorns	3.9	1.7	36.7	7.5	2.9	102.8	5.8	4.1	106.0	245.5
Colimex	6.1	1.8	38.8	10.4	2.8	137.7	10.8	4.2	158.2	334.7
Lise	7.4	1.8	53.5	8.6	2.8	88.0	7.6	3.3	127.0	268.5

L=Length of flow damage, cm; D=diameter of flow, mm; H=Eggs per flow, TH=Total of eggs in flows 1-3.

Table 5. Multiple linear regression analysis of the number of eggs (E) of *D. bulgara* in function of the variable damage length in the different flows in the five lime cultivars (FL) and the respective flow diameter (FD).

Variable	parameter			R ²	R ² adjusted	EMS	AIC	tolerance >0.10		VIF <4.0	
	β_0	β_1	β_2								
DL y FD	-55.5**	11.4**	24.7**	0.58	0.57	3100.3	1433.9	0.9	0.9	1.0	1.0
Modelo	Huevecillos = -55.569 + 11.437*LD + 24.75*DF										

EMS=Error mean square; LD=Damage length; FD=flow diameter; VIF=Variance inflation factor.

The number of fruits damaged indirectly by the effect of oviposition of the cicadas was variable between the lime cultivars evaluated and the density established. The largest number of mummified fruits (15.6) was observed in trees of the variety Colimex in productive stage (density of 312 trees ha⁻¹); while the lowest number was observed in the variety Lise in density of 312 trees ha⁻¹ (5.25 mummified fruits) (Table 6). Data from the estimation of fruit loss suggest that up to 146 kg ha⁻¹ could be lost in the variety Colimex from the indirect effect of oviposition by the cicada (Table 6).

Previous studies have reported that different species of cicadas affect different plant species. In Canada, the cicadas of long cycle of 13 to 17 years (*Magicicada* spp.) cause damage on the apical branches of young oaks, provoking a reduction of 30% of total biomass (Koenig and Liebhold, 2003). In Spain, cicadas of the genus *Cicada* sp. are considered secondary pests of olive trees, since they affect the apical branches of young plants and the nymphs feed off the roots (González *et al.*, 1998). Logan *et al.* (2014) report that the cicada *Amphipsalta zelandica* (Boisduval) in New Zealand rose as an emergent pest in kiwi crops since the year 2000, causing the same type of effects in branches and roots. On the other hand, in Brazil, various studies have been conducted of the damage caused by the common giant cicada *Quesada gigas* (Olivier), which is considered to be one of the main pests in coffee, found in 87% of the total cover of coffee plantations (Kubota, 2013).

The damage caused by cicadas happens in two different phases during the life cycle of insects. In their first stages, nymphs feed off the sap that they extract from the roots; then, in their adult stage, females cause lesions on the youngest branches during oviposition, and these branches later wither and die (Zanuncio *et al.*, 2004; Sánchez-García *et al.*, 2019). In this sense, the correct taxonomic identification of the insect, as well as the recognition of the duration of the life cycle are two important factors to develop an integral management plan (Kubota, 2013; Logan *et al.*, 2014). No control methods are reported in Mexico for this insect, although various authors have suggested some alternatives for its management. González *et al.* (1998) recommend two alternatives for the control of *Cicada* sp. in olive crops: the use of natural enemies such as the mite *Pyemotes ventricosus* (Newport) which has a depredation rate of 82% on first-instar nymphs of cicadas; and the use of lure branches or sticks such as sunflower stems or soft wood, with the aim of promoting oviposition in these sites and not in the crop of interest. In the case of damage caused by *Q. gigas* in coffee, the use of a plastic filling or plant cover traps has been recommended, with the aim of breaking

Table 6. Number of lime fruits damaged indirectly by oviposition of *D. bulgara* and estimation of fruit loss in kg ha⁻¹.

Cultivar	Distance (m)	mummified fruits (number)	Loss of fruits (kg ha ⁻¹)
Colimex	8×4	15.60	146.25
	7×4	13.85	129.40
	6×4	10.90	142.11
Lise	8×4	5.25	54.22
	7×4	6.43	69.03
	6×4	6.68	70.69

the life cycle, whether by preventing nymphs from reaching the ground or avoiding adults from performing the nuptial flight, in a similar way as other insect pests that need the ground to complete their cycle (Zanuncio *et al.*, 2004; Coria-Ávalos and Ayala-Sánchez, 2010).

On the other hand, it has been mentioned that the promotion of monocrop has favored the excessive growth of pests or the emergence of secondary pests that had not been reported before. This phenomenon is common in insects with great ability to move through the air and with a massive reproduction strategy, as is the case of cicadas (Reis *et al.*, 2002). Presently, there are few cases reported of secondary or later invasions from a species of cicada on a new host with grave repercussions. In 2002, in Brazil, an infestation of *Q. gigas* was reported in a reforested area only with trees of the species *Schizolobium amazonicum* (Huber), affecting 15 ha (Zanuncio *et al.*, 2004). Sánchez-García *et al.* (2019) reported for the first time the lime *C. aurantifolia*, as well as other citrus species and wild plants, as hosts of *D. bulgara* in Mexico. These new records could represent new cases of colonization of different hosts by cicadas present in the country; however, various studies about the biology of the cicada are still necessary.

CONCLUSIONS

In this study, the direct damage caused by the oviposition process of cicadas on the terminal branches of five lime cultivars was quantified. The results show the quantification of the indirect damage by the insect through the estimation of the fruits lost during oviposition; thus, future studies could deduce the economic loss caused by this insect. Finally, to be able to establish a program for Integral Pest Management (IPM) focused on *D. bulgara* in citrus, it is necessary to understand first the biological cycle, then to estimate the damage caused, and based on this, to develop an adequate control plan for citrus in the region of the Coast of Oaxaca.

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