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***Bacillus velezensis* LP16S – a Potential Entomopathogen for Southern Green Stink Bug, *Nezara viridula* (L.)¹**

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Abstract. Strains of *Bacillus velezensis* have been successful biological control agents for various plant diseases. A novel strain – *Bacillus velezensis* LP16S – was recently shown to inhibit mycelial growth and spore germination in four major pathogens of sorghum, *Sorghum bicolor* (L.) Moench. Southern green stink bug, *Nezara viridula* (L.), infests sorghum and has beneficial endosymbiotic bacteria in the insect gut. Because *B. velezensis* LP16S was detrimental to propagation of certain fungi, we hypothesized that *B. velezensis* LP16S might affect internal microbiota, thereby affecting survival and longevity of *N. viridula*. Adult male and female *N. viridula* were fed for 2 days green bean sections that were either sterile or contaminated with a derivative strain of *B. velezensis* LP16S (i.e., *B. velezensis* LP16SR). Diet significantly affected survival of the stink bugs. Mean survival of adults fed *B. velezensis* LP16SR (15.79 days) was significantly less than adults fed sterile beans (18.58 days). Interaction of Diet by Sex was significant, indicating that females fed *B. velezensis* LP16SR had significantly shorter mean survival (approximately 15 days) than did females fed sterile beans (approximately 21 days). Also, the former group had maximum longevity at 72 days while the latter group had maximum longevity of 92 days. From observed differences in mean survival and longevity, implications and future research should consider *B. velezensis* LP16S as a potential entomopathogen to control *N. viridula*.

Introduction

Multiple strains of *Bacillus velezensis* have been used as successful biological control agents for various plant diseases (e.g., Brannen and Kenney 1997, Ye et al. 2018 and references therein). Prom et al. (2017) identified a novel strain of *Bacillus* sp. that inhibited development of mycelial growth and spore germination in four pathogens (i.e., *Fusarium thapsinum* Klittick, Leslie, Nelson et al., Manasas, *Colletotrichum sublineola* Henn. Ex Sacc. & Trotter, *Curvularia lunata* (Wakk.) Boedijn, and *Bipolaris* spp.) of sorghum (*Sorghum bicolor* (L.) Moench). Medrano and Prom (2022) generated the whole genome of this novel strain and categorized the bacterium as *Bacillus velezensis* LP16S. Isakeit et al. (2021) demonstrated the

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efficacy of *B. velezensis* LP16S ([//www.ncbi.nlm.nih.gov/nuccore/1778620474](http://www.ncbi.nlm.nih.gov/nuccore/1778620474)) in reducing anthracnose and improving yield over non-treated plants in sorghum fields.

Southern green stink bug, *Nezara viridula* (L.) (Hemiptera: Pentatomidae), is a common pest of sorghum in Central Texas (Hall and Teetes 1981). *N. viridula* also can be a pest of cotton (*Gossypium hirsutum* L.), alfalfa (*Medicago sativa* L.), and soybean (*Glycine max* (L.) Merr.), as well as other high-value crops (e.g., fruit and nuts) (Esquivel et al. 2018 and references therein). *N. viridula* feed on bolls and transmit disease-causing pathogens of cotton (Medrano et al. 2007, 2009). Related pentatomid species can acquire gut symbionts from the environment (Kikuchi et al. 2007). Disease-causing pathogens in field-collected *N. viridula* suggest the insect can also acquire pathogens from the environment (Bell et al. 2005). *N. viridula* also harbors innate bacterial endosymbionts in the alimentary canal that are critical for life functions of the insect (Goodchild 1966, Hirose et al. 2006, Prado et al. 2006, Prado and Almeida 2009). For example, newly-hatched nymphs of *N. viridula* probe egg surfaces to acquire beneficial endosymbionts, but surface sterilization (i.e., removal of endosymbionts) of *N. viridula* eggs results in compromised fitness of emergent insects (Prado et al. 2006, Tada et al. 2011).

Because *B. velezensis* LP16S can inhibit growth of fungal pathogens of sorghum (Prom et al. 2017), we hypothesized *B. velezensis* LP16S could also adversely affect the innate endosymbiont profile of *N. viridula* and presumably affect life parameters (e.g., shortened survival / longevity of adults). As a proof-of-concept, we undertook initial efforts to examine effects of *B. velezensis* LP16S on survival and longevity of adult *N. viridula*. Results are presented with discussion focused on potential use of *B. velezensis* LP16S as a potential entomopathogen for *N. viridula*.

Materials and Methods

A colony of *N. viridula* was maintained at the USDA, ARS, Insect Control and Cotton Disease Research Unit laboratory. All colony adults and immature stages were fed a mixture of maize (*Zea mays* L.) and green beans (*Phaseolus vulgaris* L.). The green beans were rinsed in 10% sodium bicarbonate solution before feeding, providing a presumably disease-free food source. All adults were kept in BugDorm-1 containers (Mod. No. 1452, 299 cm³; BioQuip Products, Rancho Dominguez, CA), and provisioned with two ears of maize (with shuck stripped to final layer) and approximately 12 pods of green beans per container. The food items were replaced twice per week at 3- to 4-day intervals. Also, at each food replacement, available egg clutches were harvested and placed in modified plastic containers as previously described (Esquivel and Medrano 2012). Nymphs resulting from the egg clutches and colony insects were held in a walk-in environmental chamber at a photoperiod of 14:10 light:dark hours and constant 26.7 ± 1°C. The diet was replaced 2-3 times per week. Adults from the field were occasionally added to enhance hybrid vigor of the colony.

Bacillus velezensis LP16S was originally isolated from half-strength potato dextrose agar medium plated with kernels from sorghum panicles harvested at the Texas AgriLife Research Farm in Burleson County, TX, by one of the co-authors (LKP). Genome sequencing of *Bacillus velezensis* LP16S was previously reviewed, annotated, and released internationally (BioSample SAMN11373588; <https://www.ncbi.nlm.nih.gov/biosample/SAMN11373588> accessed 11 February 2022), and raw sequencing data were deposited as accession number SRX5801078 in the NCBI Sequence Read Archive database (<https://www.ncbi.nlm.nih.gov/sra/?>

term=srx5801078 accessed 11 February 2022 (Medrano and Prom 2022). The Whole-Genome Shotgun project was deposited as accession number SSKM00000000.1 at DDBJ/ENA/GenBank (<https://www.ncbi.nlm.nih.gov/nuccore/sskm00000000.1> accessed 11 February 2022).

A derivative strain of *B. velezensis* LP16S with rifampicin resistance, hereafter referred to as *B. velezensis* LP16SR, was generated using methods described by Medrano and Bell (2007). Rifampicin resistance allowed detection of the known pathogen (i.e., *B. velezensis* LP16S) when examining study insects for *B. velezensis* LP16SR (EGM, unpublished data; planned for a subsequent report). Isolated colonies of *B. velezensis* LP16SR were routinely propagated on Luria Bertani agar (LBA; Difco Laboratories, Detroit, MI) amended with rifampicin (LBAR; 100 µg ml⁻¹) at 27°C. Survival and longevity of *N. viridula* adults were assessed using a diet of green beans sterilized or contaminated with *B. velezensis* LP16SR (following paragraph).

Fifteen to 25 fresh green bean pods were sterilized 20 minutes by heating at 121°C at 1 kg cm² pressure using an autoclave (Consolidate Sterilizer Systems, Boston, MA), and cross-sectioned into pieces (approximately 3 cm) using a sterile scalpel (bean ends were discarded). To test for sterility, samples from each batch processed were triturated using a sterile mortar and pestle with 1 ml of water, plated on LBA and potato dextrose agar (PDA) (Difco Laboratories, Detroit, MI), and incubated for no longer than 2 weeks at 28°C (Medrano et al. 2007). Some of the remaining sterile beans were the 'sterile green bean' treatment, while the remainder of the beans were contaminated with *B. velezensis* LP16SR.

To generate green beans contaminated with *B. velezensis* LP16SR, suspensions of *B. velezensis* LP16SR were prepared in sterile water from 18-hour cultures (at 27°C and 12:12 [L:D] adjusted spectrophotometrically ($A_{600} = 1.0$), resulting in a concentration of 10¹¹ cells ml⁻¹. The inoculum for the remaining green beans was prepared by adding 1 ml of *B. velezensis* LP16SR suspension to 49 ml of sterile water, resulting in an inoculum concentration of 2 × 10⁹ cells ml⁻¹.

After sterilization, green bean sections provided to insects were soaked for 2 minutes in sterile water or *B. velezensis* LP16SR inoculum, blotted dry using sterile paper towels, and put individually in a sterile, disposable Petri dish (100 × 15 mm; VWR, Missouri City, TX). A single insect of determined sex was put in a Petri dish containing a sterile green bean section or a section contaminated with *B. velezensis* LP16SR.

Individual laboratory-reared, newly-eclosed (<24 hours old) *N. viridula* adults were used as experimental units. To obtain newly-eclosed adults, 5th instars were selected from the colony, put in BugDorm-1 containers, and fed colony diet as described previously. As newly-eclosed adult *N. viridula* became available, individual adults were sexed and put in a respective Petri dish. Each Petri dish contained a sterilized green bean section (as checks) or green bean section contaminated with *B. velezensis* LP16SR (described previously) to assess the influence of *B. velezensis* LP16S on adult survival and longevity. Each insect was exposed for 2 days to a diet of a green bean section sterilized or contaminated with *B. velezensis* LP16SR. At the conclusion of the 2-day feeding period, flame-sterilized forceps were used to avoid cross-contamination and each adult was transferred to a respective new Petri dish containing a single, non-sterile, whole green bean that had been rinsed as previously described. Beans were replaced every Monday, Wednesday, and Friday until an insect died. Petri dishes were replaced if very soiled. Survival (days) was recorded daily, and numbers of dead males and females were recorded daily. Study

insects were kept in an environmental chamber (Model I30-BLL; Percival Scientific, Percy, IA) at photoperiod of 14:10 light:dark hours and constant $29.4 \pm 1^\circ\text{C}$.

Four replications were used to assess the influence of *B. velezensis* LP16S (by the derived strain) on survival and longevity of *N. viridula*. A replication had ≥ 50 *N. viridula* adults per treatment, and the number of southern green stink bug adults per replication ranged from 101 (Replication 1) to 120 (Replication 4) (Table 1). Approximately a 50% sex ratio was maintained, except Replication 3 because of insufficient newly-eclosed males.

Influence of *B. velezensis* LP16S on survival of *N. viridula* adults was analyzed using a generalized linear mixed model (GLMM) described by Stroup (2013). PROC GLIMMIX (SAS Institute Inc., 2016, Version 9.4, SAS/STAT 15.1, Cary, NC) with appropriate options (e.g., inclusion of random model effects) was previously shown to statistically model time-to-event data in entomological studies (Yeater et al. 2019, Yocom et al. 2019). For current *N. viridula* survival data, GLMM by the GLIMMIX procedure is preferred to obtain estimates of survivor and hazard functions. The dependent variable was observed time to death (expressed as days) after exposure to Diet. Fixed effects in the model were Diet treatments (i.e., exposure to green bean section sterilized or contaminated with *B. velezensis* LP16SR), Sex, and their interaction. Replication effect was considered random. Time-to-event data were not normally distributed; gamma and exponential distributions were assessed to determine a better fit. As described in Stroup (2013), the confidence interval for the scale parameter did not contain 1, therefore the gamma distribution was assumed, and the gamma model was fit using the DIST=GAMMA option in the MODEL statement. Additionally, Laplace's approximation (METHOD=LAPLACE in the PROC GLIMMIX statement) was applied because of conditional log-likelihood in the gamma distribution, and G-side random effects were assumed to be normal ($0, \sigma^2$). The LSMEANS statement was used to generate LSMEANS and standard errors for fixed effects. In the LSMEANS statement, the PDIFF ADJUST =TUKEY option was used to invoke the Tukey-Kramer adjustment for *post hoc* multiple pairwise comparisons of LSMEANS. The LINES option was used to visualize separation of pairwise comparisons with letter groupings ($\alpha = 0.05$). In the LSMEANS statement, the ILINK option was used to calculate back-transformed means and their standard errors.

Estimated hazard functions were calculated by dividing 1 by back-transformed mean value ($h_{ij}(t) = \lambda_{ij}$) for each Diet-by-Sex combination. A survivor function ($S(t)$) was calculated for each Diet-by-Sex combination during the length of time for the experiment (0-95 days), where $S_{ij}(t) = e^{-\lambda_{ij}t}$ (i.e., exp-time x hazard estimate). The plot of survivor functions for each Diet-by-Sex interaction was based on these estimates.

Table 1. Number (*n*) of Female and Male *N. viridula* Adults per Replication Exposed to Sterile Green Beans or *B. velezensis* LP16SR-contaminated Beans

Replication	Treatment of beans	Sex		
		Female (<i>n</i>)	Male (<i>n</i>)	Total (<i>n</i>)
1	<i>B. velezensis</i> LP16SR-contaminated	25	26	51
	Sterile	24	26	50
2	<i>B. velezensis</i> LP16SR-contaminated	30	29	59
	Sterile	30	29	59
3	<i>B. velezensis</i> LP16SR-contaminated	36	23	59
	Sterile	36	23	59
4	<i>B. velezensis</i> LP16SR-contaminated	31	29	60
	Sterile	29	31	60

Results

Diet treatment regime significantly affected mean insect survival ($F = 4.66; 1, 410; P = 0.0315$). Adults fed green bean sections contaminated with *B. velezensis* LP16SR had significantly shorter mean (LSMeans) survival of 2.76 ± 0.07 days ($= 15.79 \pm 1.08$ days, back-transformed mean), whereas adults fed sterile green beans had greater mean survival of 2.92 ± 0.07 days ($= 18.58 \pm 1.26$ days, back-transformed mean). Sex of insect was not a significant factor affecting mean insect survival ($F = 0.35; 1, 410; P = 0.5540$) and will not be expounded upon considering the information was uninformative because of the nature of analyses (i.e., this factor pooled insects across Diet treatments).

The interaction term of Diet-by-Sex was a significant factor affecting mean insect survival ($F = 5.11; 1, 410; P = 0.0243$). Pair-wise comparisons of Diet-by-Sex combinations indicated only females fed *B. velezensis* LP16SR-contaminated beans had significantly less mean (LSMeans) survival than females fed sterile beans (Table 2; $Adj. P = 0.0083$). Back-transformed mean survival for females fed *B. velezensis* LP16SR-contaminated beans was approximately 15 days, while mean survival for females fed sterile beans was approximately 21 days. No remaining pair-wise means (LSMeans) comparisons were significantly different ($0.1853 \leq Adj. P \leq 1.000$).

Survival function curves plotting the hazard function estimates for each Diet treatment by Sex interaction further demonstrate differences in calculated survival over time (Fig. 1). Fifty percent survival of females was at approximately 10 and 14 days after exposure to *B. velezensis* LP16SR-contaminated beans and sterile green beans, respectively. Ninety percent mortality of females was at 34 and 47 days after exposure to *B. velezensis* LP16SR-contaminated beans and sterile green beans, respectively. The survival curve for males fed *B. velezensis* LP16SR-contaminated beans essentially mirrored that of males fed sterile beans.

Discussion

Bacillus velezensis LP16SR detrimentally affected survival and longevity of female *N. viridula*. *Nezara viridula* possess innate beneficial endosymbiotic microbes (Hirose et al. 2006, Prado et al. 2006, Prado and Almeida 2009, Prado and Zucchi 2012). A compromised innate endosymbiont system resulting from sterilization of egg surfaces can affect fitness of resultant *N. viridula* nymphs and adults (Prado et al. 2006, Tada et al. 2011). Similarly, fitness effects were observed in *Halyomorpha*

Table 2. Observed Mean Survival (days; LSMeans and back-transformed means, with standard errors [SE]) and Longevity (i.e., range) of *N. viridula* Adults Exposed to Sterile Green Bean Sections or *B. velezensis* LP16SR-contaminated Beans

Treatment of beans	Sex	n	LSMeans	Back-transformed	Range (days)
			Estimate* \pm SE	Mean \pm SE	
Sterile	♀♀	108	$3.02a \pm 0.08$	20.66 ± 1.75	2-92
Sterile	♂♂	100	$2.82ab \pm 0.09$	16.72 ± 1.45	1-55
<i>B. velezensis</i> LP16SR	♀♀	110	$2.70b \pm 0.08$	14.85 ± 1.25	1-72
<i>B. velezensis</i> LP16SR	♂♂	99	$2.82ab \pm 0.09$	16.80 ± 1.48	1-65

*LSMeans estimates followed by the same lowercase letter are not significantly different based on Tukey-Kramer Adjusted P (where $Adj. P \leq 0.05$).

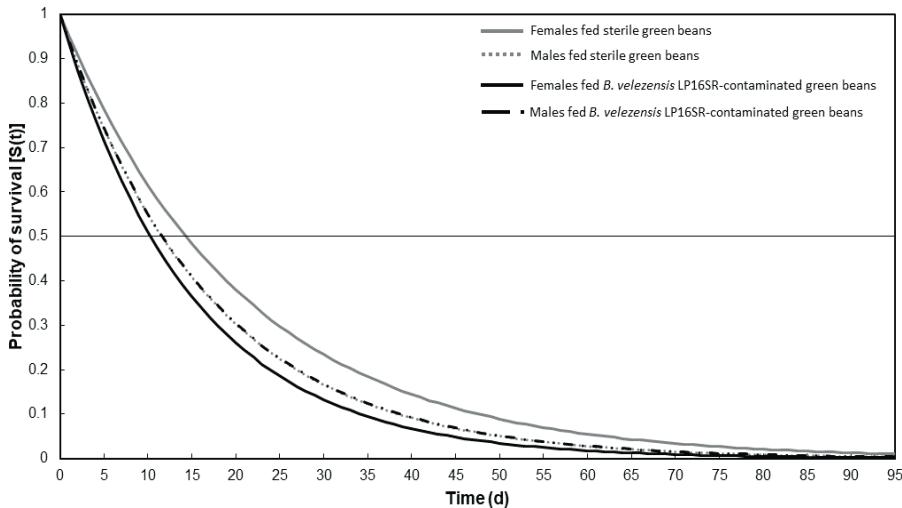


Fig. 1. Survival curves of hazard estimates $[S(t)]$ for adult *Nezara viridula* females and males fed either sterile green bean sections or *B. velezensis* LP16SR-contaminated green bean sections, where Time (d) is age in days at time of death after exposure to diet treatments. Horizontal reference line highlights 50% probability survival time after exposure to diet treatments.

halys Stål (Hemiptera: Pentatomidae) nymphs and adults resulting from eggs that were surface-sterilized (Taylor et al. 2014). The observed differences (i.e., shortened mean survival and reduced longevity) between females fed *B. velezensis* LP16SR and sterile beans have significant biological implications and suggested *B. velezensis* LP16S (via *B. velezensis* LP16SR) acted as an entomopathogen detrimentally affecting the biology of *N. viridula* females. Mechanisms involved in causing the shorter life span are not currently known, and elucidation of the mechanisms was outside the scope of the current study. Several hypotheses where *B. velezensis* LP16S has potential as an entomopathogen were proposed.

Improved understanding of the mechanism(s) producing shortened mean survival and reduced longevity in females would enhance potential use of *B. velezensis* LP16S as a biological control agent or entomopathogen. Spores of *B. velezensis* produce biofilm that protects the rhizosphere of plants from otherwise pathogenic invaders (i.e., phytopathogens), and it is believed the biofilm is the mechanism inferring biocontrol properties against phytopathogen infection (Ye et al. 2018 and references therein). *B. velezensis* LP16S mitigates fungi – primarily by inhibiting mycelial growth and spore germination of pathogens that cause diseases in sorghum (Prom et al. 2017, Isakeit et al. 2021). Plausibly, in females, observed decreased mean survival and longevity might be a function of detrimental effects to the microbiome caused by biofilm formation in the alimentary canal and, consequently, reduced fitness.

Whether additional biological functions such as feeding frequency, oviposition rate/frequency, fecundity, and hatch rate are affected has yet to be determined. A preliminary glimpse of feeding being affected can be seen in females fed *B.*

velezensis LP16SR-contaminated beans; that is, although some females survived 72 days after exposure to treated beans, frequency of feeding thereafter might have been compromised, thereby leading to observed less mean survival and reduced life span of *N. viridula*. Effects of *B. velezensis* LP16S do not have to be mutually exclusive and could show additional promise as a biological control agent if adults also fed and oviposited less often and deposited fewer eggs, resulting in reduced hatch rate. These biological parameters could be future research avenues to elucidate the impact of *B. velezensis* LP16S on the biology of *N. viridula*. Such efforts would align with the idea posed by Prado and Zucchi (2012) that "...manipulation of symbiotic interactions can contribute to the development of new strategies for pest control, including...insects...". In the current work, *B. velezensis* LP16S might similarly be manipulating the symbiotic relationship of innate bacteria by negatively interfering with innate endosymbionts and their expected roles in maintaining healthy, vigorous, and productive adults.

Ruiz-Garcia et al. (2005) originally isolated and described *B. velezensis* from the River Vélez in southern Spain. The taxonomic status of *B. velezensis* was recently amended with the conclusion that "...*B. methylotrophicus* KACC 13015^T, *B. amyloliquefaciens* subsp. *plantarum* FZB42^T, and '*B. oryzicola*' KACC 18228 should be reclassified as later heterotypic synonyms of *B. velezensis* NRRL B-41580^T, since the valid publication date of *B. velezensis* precedes the other three strains." (Dunlap et al. 2016). The literature is rife with examples of *Bacillus* spp. as effective biological control agents against fungal pathogens [e.g., see Jacobsen et al. 2004 and references therein] but few records were located related to *Bacillus velezensis* affecting insect behavior (i.e., Rashid et al. 2017, 2018; Liang et al. 2022). *B. velezensis* was detected in another pest pentatomid – *Piezodorus guildinii* (Westwood) – but the mechanism for acquisition of *B. velezensis* (or its effects on the insect) was not determined (Husseneder et al. 2017).

Comparing our findings with Rashid et al. (2017, 2018), our results are the first record of *B. velezensis* exhibiting potential insect pest control by direct detrimental effects of *B. velezensis* LP16S on the biology of *N. viridula* (i.e., reduced stink bug survival and longevity). That is, in evaluating *B. velezensis* YC7010 (as *Bacillus oryzicola* YC7010) against the brown planthopper (*Nilaparvata lugens* Stål [Hemiptera: Delphacidae]) and the green peach aphid (*Myzus persicae* (Sulzer) [Hemiptera: Aphididae]) Rashid et al. (2017, 2018) observed "systemic resistance" caused by metabolic (e.g., production of secondary metabolites) and physical changes (i.e., "...strengthening of physical barriers...") in the plants leading to behavioral changes in insects (e.g., less feeding frequency). Effectively, these earlier reports demonstrated antixenotic effects toward insects (i.e., changes to plant properties affected the behavior of *M. persicae* and *N. lugens*) caused by plants incorporating *B. velezensis* YC7010. Conversely, our results demonstrated direct antibiosis (or insecticidal) effects toward insects (i.e., direct deleterious effect on insect biology) caused by *N. viridula* exposure and presumed ingestion of *B. velezensis* LP16SR. Similarly, a recombinant strain of *B. velezensis* resulted in "...high insecticidal activity against a lepidopterous pest, *Plutella xylostella*..." but this recombinant strain required modification of *B. velezensis* S3-5 by incorporating the Cry1Ac crystal protein found in *Bacillus thuringiensis* (Roh et al. 2009). Rabbee et al. (2019) provide a comprehensive review of genes and secondary metabolites involved in biological control properties of *B. velezensis* toward plant diseases. Efforts are planned to conclusively decipher the relationship between the biology of *N. viridula* and pathogen ingestion.

Our results offer proof-of-concept that *B. velezensis* LP16S could be a potential entomopathogen for *N. viridula*. Should *B. velezensis* LP16S prove to successfully affect other reproductive parameters of *N. viridula* adults, the practicality and delivery method of the entomopathogen to field populations of *N. viridula* need to be determined. In a review of *Bacillus* spp. as biological control agents, Jacobsen et al. (2004) mentioned spray applications of *Bacillus* spp. to mitigate crop diseases. Isakeit et al. (2021) sprayed *B. velezensis* LP16S on sorghum plants and found the plants yielded more and had less disease than non-treated sorghum plants; presumably, *B. velezensis* LP16S reduced colonization of fungal pathogens in sorghum. Thus, Isakeit et al. (2021) established precedence for improved benefits and delivery method of *B. velezensis* LP16S to sorghum.

N. viridula is a pest of sorghum, and adults oviposit in sorghum (Tillman 2013). Exploitation of feeding behavior of 1st instars of *N. viridula* might provide additional opportunity to evaluate *B. velezensis* LP16S as an entomopathogen. Specifically, soon after hatching, 1st instars of *N. viridula* probe the egg chorion to acquire crucial endosymbionts required for a healthy insect to thrive. Esquivel and Medrano (2020) demonstrated that 1st instars also feed and acquire a cotton pathogen (i.e., *Pantoea agglomerans* (Ewing and Fife)), subsequently retaining the pathogen until adults. Application of *B. velezensis* LP16S on available egg clutches might provide a mechanism to introduce the entomopathogen into 1st instars and subsequent stadia. Should 1st instars also ingest *B. velezensis* LP16S, the agent could hypothetically cause mortality before nymphs become reproductive adults.

Combined with results here examining the influence of *B. velezensis* LP16S on shortened mean adult survival and reduced longevity, potential research examining adult bionomics and ingestion by nymphs could further support the idea of *B. velezensis* LP16S as an entomopathogen for *N. viridula*. Our current findings confirm antibiosis of *B. velezensis* LP16S on *N. viridula*, and research is readily identified to further elucidate interactions of the bacterium with life processes of *N. viridula*.

References Cited

- Bell, A. A., J. Lopez, J. Esquivel, E. Medrano, and J. Mauney. 2005. Isolation of cottonseed-rotting *Pantoea* spp. from stink bugs and plant bugs, pp. 186-193. In Proceedings of the 2005 Beltwide Cotton Conferences, New Orleans, LA. National Cotton Council, Memphis, TN.
- Brannen, P. M., and D. S. Kenney. 1997. Kodiak®—a successful biological-control product for suppression of soil-borne plant pathogens of cotton. J. Ind. Microbiol. Biotechnol. 19: 169-171. <https://doi.org/10.1038/sj.jim.2900439>.
- Dunlap, C. A., S.-J. Kim, S.-W. Kwon, and A. P. Rooney. 2016. *Bacillus velezensis* is not a later heterotypic synonym of *Bacillus amyloliquefaciens*; *Bacillus methylotrophicus*, *Bacillus amyloliquefaciens* subsp. *plantarum* and '*Bacillus oryzicola*' are later heterotypic synonyms of *Bacillus velezensis* based on phylogenomics. Int. J. Syst. Evol. Microbiol. 66: 1212-1217.
- Esquivel, J. F., and E. G. Medrano. 2012. Localization of selected pathogens of cotton within the southern green stink bug. Entomol. Exp. Appl. 142: 114-120.
- Esquivel, J. F., and E. G. Medrano. 2020. Retention of *Pantoea agglomerans* Sc1R across stadia of the southern green stink bug, *Nezara viridula* (L.) (Hemiptera: Pentatomidae). PLoS ONE 15: e0242988. <https://doi.org/10.1371/journal.pone.0242988>.

- Esquivel, J. F., D. L. Musolin, W. A. Jones, W. Rabitsch, J. K. Greene, M. D. Toews, C. F. Schwertner, J. Grazia, and R. M. McPherson. 2018. *Nezara viridula* (L.), pp. 331-403. In J. E. McPherson [ed.], Invasive Stink Bugs and Related Species (Pentatomidae): Biology, Higher Systematics, Semiochemistry, and Management. CRC Press, Boca Raton, FL.
- Goodchild, A. J. P. 1966. Evolution of the alimentary canal in the Hemiptera. Biol. Rev. 41: 97-140.
- Hall, D. G., IV, and G. L. Teetes. 1981. Alternate host plants of sorghum panicle-feeding bugs in southeast Central Texas. Southwest. Entomol. 6: 220-228.
- Hirose, E., A. R. Panizzi, J. T. De Souza, A. J. Cattelan, and J. R. Aldrich. 2006. Bacteria in the gut of southern green stink bug (Heteroptera: Pentatomidae). Ann. Entomol. Soc. Am. 99: 91-95.
- Husseneder, C., J.-S. Park, A. Howells, C. V. Tihke, and J. A. Davis. 2017. Bacteria associated with *Piezodorus guildinii* (Hemiptera: Pentatomidae), with special reference to those transmitted by feeding. Environ. Entomol. 46: 159-166.
- Isakeit, T., S. Labar, W. L. Rooney, and L. K. Prom. 2021. Effect of *Bacillus velezensis* and fungicides on sorghum anthracnose in Burleson County, Texas, 2020. Plant Dis. Management Reports 15: CF076.
- Jacobsen, B. J., N. K. Zidack, and B. J. Larson. 2004. The role of *Bacillus*-based biological control agents in integrated pest management systems: plant diseases. Phytopath. 94: 1272-1275.
- Kikuchi, Y., T. Hosokawa, and T. Fukatsu. 2007. Insect-microbe mutualism without vertical transmission: a stinkbug [sic] acquires a beneficial gut symbiont from the environment every generation. Appl. Environ. Microbiol. 73: 4308-4316. <https://doi.org/10.1128/AEM.00067-07>
- Liang, L., Y. Fu, S. Deng, Y. Wu, and M. Gao. 2022. Genomic, antimicrobial, and aphicidal traits of *Bacillus velezensis* ATR2, and its biocontrol potential against ginger rhizome rot disease caused by *Bacillus pumilus*. Microorganisms 10: 63. <https://doi.org/10.3390/microorganisms10010063>
- Medrano, E. G., and L. K. Prom. 2022. Genome sequence data of *Bacillus* sp. strain LP16S which is capable of inhibiting the growth of multiple sorghum fungal pathogens. MPMI [Molecular Plant-Microbe Interactions] 35: 290-292. <https://doi.org/10.1094/MPMI-10-21-0246-A>
- Medrano, E. G., J. F. Esquivel, and A. A. Bell. 2007. Transmission of cotton seed and boll rotting bacteria by the southern green stink bug (*Nezara viridula* L.). J. Appl. Microbiol. 103: 436-444.
- Medrano, E. G., J. Esquivel, A. Bell, J. Greene, P. Roberts, J. Bacheler, J. Marois, D. Wright, and R. L. Nichols. 2009. Potential for *Nezara viridula* (Hemiptera: Pentatomidae) to transmit bacterial and fungal pathogens into cotton bolls. Curr. Microbiol. 59: 405-412. doi:10.1007/s00284-009-9452-5.
- Prado, S. S., and R. P. P Almeida. 2009. Phylogenetic placement of pentatomid stink bug gut symbionts. Curr. Microbiol. 58: 64-69.
- Prado, S. S., and T. D. Zucchi. 2012. Host-symbiont interactions for potentially managing heteropteran pests. Psyche Vol. 2012, Article ID 269473. DOI: 10.1155/2012/269473
- Prado, S. S., D. Rubinoff, and R. P. P. Almeida. 2006. Vertical transmission of a pentatomid caeca-associated symbiont. Ann. Entomol. Soc. Am. 99: 577-585.
- Prom, L. K., E. G. Medrano, T. Isakeit, R. Jacobsen, and R. Droleskey. 2017. A pictorial illustration of the inhibition of mycelial growth and spore germination

- of various sorghum fungal pathogens by a *Bacillus* species [sic]. Res. J. Plant Pathol. 1: 1-5.
- Rabbee, M. F., M. S. Ali, J. Choi, B. S. Hwang, S. C. Jeong, and K. Baek. 2019. *Bacillus velezensis*: a valuable member of bioactive molecules within plant microbiomes. Molecules 24: 1046.
- Rashid, M. H., A. Khan, M. T. Hossain, and Y. R. Chung. 2017. Induction of systemic resistance against aphids by endophytic *Bacillus velezensis* YC7010 via expressing PHYTOALEXIN DEFICIENT4 in *Arabidopsis*. Front. Plant Sci. 8: 211. DOI: 10.3389/fpls.2017.00211.
- Rashid, M. H., H.-J. Kim, S.-I. Yeom, H. A. Yu, M. M. Manir, S.-S. Moon, Y. J. Kang, and Y. R. Chung. 2018. *Bacillus velezensis* YC7010 enhances plant defenses against brown planthopper through transcriptomic and metabolic changes in rice. Front. Plant Sci. 9: 1904. <https://doi.org/10.3389/fpls.2018.01904>
- Roh, J. Y., Q. Liu, J. Y. Choi, Y. Wang, H. J. Shim, H. G. Xu, G. J. Choi, J. C. Kim, and Y. H. Je. 2009. Construction of a recombinant *Bacillus velezensis* strain as an integrated control agent against plant diseases and insect pests. J. Microbiol. Biotechnol. 19: 1223-1229. DOI: 10.4014/jmb.0902.065.
- Ruiz-García, C., V. Béjar, F. Martínez-Checa, I. Llamas, and E. Quesada. 2005. *Bacillus velezensis* sp. nov., a surfactant-producing bacterium isolated from the river Vélez in Málaga, southern Spain. Int. J. Syst. Evol. Microbiol. 55: 191-195. <https://doi.org/10.1099/ijst.0.63310-0>
- Stroup, W. W. 2013. Generalized Linear Mixed Models: Modern Concepts, Method and Applications, pp. 375-395. CRC Press, Boca Raton, FL.
- Tada, A., Y. Kikuchi, T. Hosokawa, D. L. Musolin, and K. Fujisaki. 2011. Obligate association with gut bacterial symbiont in Japanese populations of the southern green stinkbug [sic, no space between stinkbug] *Nezara viridula* [sic, no punctuation before/after scientific name] (Heteroptera: Pentatomidae). Applied Entomology and Zoology 46: 483-488. DOI 10.1007/s13355-011-0066-6
- Taylor, C. M., P. L. Coffey, B. D. DeLay, and G. P. Dively. 2014. The importance of gut symbionts in the development of the brown marmorated stink bug, *Halyomorpha halys* (Stål). PLoS ONE 9: e90312. DOI: 10.1371/journal.pone.0090312.
- Tillman, G. P. 2013. Stink bugs (Heteroptera: Pentatomidae), a leaffooted bug (Hemiptera: Coreidae), and their predators in sorghum in Georgia. J. Entomol. Sci. 48: 9-16.
- Ye, M., X. Tang, R. Yang, H. Zhang, F. Li, F. Tao, F. Li, and Z. Wang. 2018. Characteristics and application of a novel species of *Bacillus*: *Bacillus velezensis*. ACS Chem. Biol. 13: 500-505. DOI: 10.1021/acscchembio.7b00874.
- Yeater, K., G. Yocom, K. Greenlee, J. Bowsher, A. Rajamohan, and J. Rinehart. 2019. Generalized linear mixed model approach to time-to-event data with censored observations. Southeast SAS Users Group Conference Paper 160-2019. <https://par.nsf.gov/biblio/10163553>.
- Yocom, G. D., J. P. Rinehart, A. Rajamohan, J. H. Bowsher, K. M. Yeater, and K. J. Greenlee. 2019. Thermoprofile parameters affect survival of *Megachile rotundata* during exposure to low-temperatures. Integr. Comp. Biol. 59: 1089-1102. <https://doi.org/10.1093/icb/icz126>.

***Metarhizium acridum*¹ and Other Entomopathogenic Fungi from Grasshoppers² at Arid Sites of Northeastern Mexico**

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Abstract. Entomopathogenic fungi are important natural enemies of the Orthoptera, insects of great economic impact. We collected 530 live and almost 200 dead grasshoppers near Saltillo, Mexico, during 2007-2010. Less than 1% of live, incubated, and as many as 30% of dead insects were infected by *Beauveria bassiana* (Bals.) Vuill. sensu lato (in dead insects, coinfection with *Entomophaga* spp.). *Metarhizium acridum* (Driver & Milner) Bischoff, Rehner & Humber was observed in live *Schistocerca nitens* Thunberg adults. Therefore, we enumerated five species of fungi from locusts and other grasshoppers at Saltillo: besides *B. bassiana* and *M. acridum*, *Metarhizium anisopliae* (Metch.) Sorokin sensu lato (all Ascomycota: Hypocreales); and *Entomophaga calopteni* (Bessey) Humber and *Entomophaga macleodii* Humber (unpublished) (Zoopagomycota: Entomophthorales). First reported in this work were *M. acridum* from an arid zone in the Nearctic region, *S. nitens* as a host of *M. acridum* and *Entomophaga* sp. in Mexico, combined infections by *B. bassiana* and *Entomophaga* spp. on melanopline grasshoppers, and new distribution records for *E. calopteni* in Mexico. Internal sporulation of *Beauveria* on large melanopline grasshoppers was observed. The impact of these natural enemies should be verified. The Chihuahuan Desert strains of *M. acridum* should be considered for control of North American grasshopper pests.

Resumen. Los hongos entomopatógenos son importantes enemigos naturales de los ortópteros, insectos de gran impacto económico. Recolectamos 530 saltamontes vivos y cerca de 200 muertos cerca de Saltillo, México, durante 2007-2010. Menos del 1% de los insectos vivos, incubados, y hasta el 30% de los insectos muertos fueron infectados por *Beauveria bassiana* (Bals.) Vuill. sensu lato (en insectos muertos, coinfección con *Entomophaga* spp.). Se observó *Metarhizium acridum* (Driver & Milner) Bischoff, Rehner & Humber en adultos vivos de *Schistocerca nitens* Thunberg. Por lo tanto, enlistamos cinco especies de hongos de langostas y otros saltamontes en Saltillo: además de *B. bassiana* y *M. acridum*, *Metarhizium anisopliae* (Metch.) Sorok. sensu lato (Ascomycota: Hypocreales); y *Entomophaga calopteni*

¹Hypocreales: Cordycipitaceae

²Orthoptera: Acrididae

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(Bessey) Humber y *Entomophaga macleodii* Humber (sin publicar) (Zoopagomycota: Entomophthorales). Los primeros reportes en este trabajo son *M. acridum*, de una zona árida en la región Neártica, *S. nitens* como huésped de *M. acridum* y *Entomophaga* sp. en México, infecciones combinadas por *B. bassiana* y *Entomophaga* spp. en saltamontes melanoplinos, y nuevos registros de distribución de *E. calopteni* en México. Se observó esporulación interna de *Beauveria* en saltamontes melanoplinos grandes. Se debe verificar el impacto de estos enemigos naturales. Las cepas del desierto de Chihuahua de *M. acridum* deben considerarse para el control de plagas de saltamontes de América del Norte.

Introduction

Grasshoppers (Orthoptera: Acrididae, Romaleidae and other families) can cause extensive damage in agriculture. Several fungal pathogens are natural biological control agents of grasshoppers (Prior and Greathead 1989). There is worldwide interest in manipulation of fungi for control of acridids (Kooyman et al. 1997, Sanchez-Peña 2005, Kamga et al. 2022). In Mexico, grasshoppers of economic importance include the ebony grasshopper, *Boopedon nubilum* Say; many species of spur-throated grasshoppers, mostly in the genus *Melanoplus*; Central American locust, *Schistocerca piceifrons piceifrons* (Walker); corn field grasshopper, *Sphenarium purpurascens* Charpentier; plains lubber grasshopper, *Brachystola* spp., and western horse lubber grasshopper, *Taeniopoda eques* (Romaleidae) (Salas-Araiza et al. 2003, SENASICA 2022). Entomopathogenic fungi in the Hypocreales (Ascomycota) reported in Mexico infecting grasshoppers are: *Metarhizium acridum* (Driver & Milner) Bischoff, Rehner & Humber (= *Metarhizium anisopliae* var. *acridum* Driver, Milner & Trueman; some strains of *Metarhizium flavoviride* W. Gams & Rozsypal also belong in this species). *M. acridum* was found infecting *S. piceifrons* in the tropical western coastal region of the Pacific (Hernández-Velázquez et al. 2003). Also, in the arid and semi-arid north, *Beauveria* cf. *bassiana* (Bals.) Vuill. and *Metarhizium* cf. *anisopliae* (Metch.) Sorok. were collected from *Brachistola magna* (Girard) in Durango and Chihuahua states (Barajas-Ontiveros et al. 2009, García-Pereyra et al. 2014). Acridid-pathogenic fungi in the genus *Entomophaga* (Zoopagomycota: Entomophthorales), formerly known as *Entomophthora grylli* Fres. and *Entomophaga grylli* (Fres.), are a species complex. An epizootic of an unidentified species of *Entomophaga* killed *Mermiria bivittata* in southern Tamaulipas State (Barrientos-Lozano et al. 2002). Two *Entomophaga* species were repeatedly observed coexisting at Saltillo, Coahuila: *Entomophaga grylli* pathotype I, informally called “*Entomophaga macleodii*” by R. A. Humber, USDA-ARS (hereafter called *E. macleodii*); and *Entomophaga calopteni* (Bessey) Humber (= *Entomophaga grylli* pathotype II) (Sanchez-Peña 2000, 2005; Torres-Acosta et al. 2010; Casique-Valdes et al. 2012). Bioinsecticidal preparations of Mexican and exotic (Australian) isolates of *M. acridum* were evaluated in the laboratory and field in Mexico (Hernández-Velázquez et al. 2003), and sporadic efforts during the last decades tried to control *S. piceifrons* and other grasshoppers. This report is a summary of published and new records on the assortment of pathogenic fungi occurring naturally on grasshoppers in northeastern Mexico (mainly arid areas near Saltillo, Coahuila) from 2007-2021.

Entomophaga spp. (Zoopagomycota: Entomophthorales) are among fungi reported on Orthoptera at Saltillo where periodical autumn epizootics have been observed since the 1990s (Sanchez-Peña 2000) until 2021. The fungi induce cosmopolitan “summit disease” of grasshoppers: a few hours before dying, infected

individuals climb to the tips of vegetation, posts, fences, and other elevated sites, holding themselves firmly with all legs onto the substrate where they die (Fig. 1). More than 20 species of Acrididae are at Saltillo, and several are hosts of the fungi (Casique-Valdés et al. 2012). *Entomophaga macleodii* mainly infects banded-winged grasshoppers (subfamily Oedipodinae), while *E. calopteni* primarily infects spur-throated grasshoppers, subfamily Melanoplinae (Ramoska et al. 1988). Only *E. macleodii* produces small, inconspicuous conidiophores and infective spores externally on the insect (Fig. 2). In *E. calopteni*, no external fungal growth is visible on infected insects (Figs. 1-2); the fungus produces only spherical, resistant chlamydospores (= resting spores) that fill the host; this occurs also in *E. macleodii*. A critical difference between the species is that *E. macleodii* can spread directly from infected to healthy individuals by conidia produced externally on killed hosts, while in *E. calopteni*, infective conidia are produced mostly from germinating resting spores in the environment (litter and soil) (Ramoska et al. 1988). Casique-Valdez et al. (2012) amplified DNA markers to identify *Entomophaga* spp. in grasshoppers from Saltillo. The most abundant host species were *Melanoplus thomasi* Scudder (reported there as *M. bivittatus*) and brachypterous *Phoetaliotes nebrascensis* (Thomas). A few hosts had mixed infections (i.e., single individual testing positive for both *Entomophaga* species); of 330 *Entomophaga*-infected grasshoppers analyzed by sequencing, mixed infections were detected in nine individuals: five *M. thomasi*, one *Phaedrotettix angustipennis* Scudder, and one



Fig. 1. "Summit disease" of grasshoppers infected by *Entomophaga calopteni*, a member of the *Entomophaga grylli* species complex, Saltillo, Mexico. Left, two adults of *Melanoplus thomasi* killed by *Entomophaga calopteni*. Infected, moribund insects climb on vegetation (in this case silverleaf nightshade, *Solanum eleagnifolium*), fences, etc. Insects die firmly attached to plants, disintegrate (i.e., individual on right), and resting spores filling the body fall to the ground and become dormant in soil. Right, cluster of *Melanoplus* spp. (mainly *M. thomasi*) killed by *E. calopteni*.

P. nebrascensis (Melanoplinae); one *Arphia pseudonietana* (Thomas) (Oedipodinae); and one *Mermiria bivittata* (Serville) (Gomphocerinae) (Fig. 3). Mixed infections by species of *Entomophaga* occurred in as many as 3% of grasshoppers at South Dakota (Bidochka et al. 1997).



Fig. 2. "*Entomophaga macleodii*" (*E. grylli* pathotype I) on *Arphia pseudonietana* (left), and *Entomophaga calopteni* (*E. grylli* pathotype II) on *Melanoplus* sp. (right). External, faint growth of conidiophores on abdomen of *A. pseudonietana* (arrows); such growth is absent on *Melanoplus*. Saltillo, Mexico, October 2007.



Fig. 3. *Mermiria bivittata* (Gomphocerinae) infected by *Entomophaga* sp., Saltillo, Mexico.

Materials and Methods

Insects were collected from August-December 2007-2010 at agricultural fields and reforestation plots of the Universidad Autonoma Agraria Antonio Narro (UAAAN), Saltillo, Coahuila, Mexico. The site is in arid Chihuahuan Desert adjacent to the Sierra Madre Oriental of northeastern Mexico, at coordinates 25.357, -101.040, and 1,600 m above sea level. Most abundant species were *M. thomasi* and brachypterous *P. nebrascensis*. Live insects (530 total) were kept in screened cages in a laboratory and fed fresh grass and forb foliage until dead. More than 200 dead grasshoppers (mostly dead in the “summit disease” position) were collected in the same location and period. Insects collected alive and dying in cages, and dead insects from the field were put into humid chambers (Petri dishes with a disk of blotting paper at the bottom, moistened with sterile water). During the same period, 60 insects collected dead in the “summit disease” position were dissected and examined microscopically for fungi. The insects did not show external fungal growth. The American locust or gray bird grasshopper, *Schistocerca nitens* (Cyrtacanthacridinae), was the dominant species of *Schistocerca* in the area, often observed as scattered, solitary individuals at Saltillo. The genus *Schistocerca* has been variously identified in the subfamilies Melanoplinae or Cyrtacanthacridinae. We collected a total of 40 live adults (both sexes) of *S. nitens* in October 2008, at the UAAAN fields. These were kept in screened cages in a laboratory and fed freshly cut grass. Conditions in the laboratory were 25-28°C and 25-40% relative humidity. Vegetation also was searched for dead *S. nitens*. Dead grasshoppers with fungal infections were also searched for at the municipality of Galeana, NL, Mexico, in October-November 2021.

Results

Less than 1% of 530 grasshoppers collected live from 2007-2010 showed *B. cf. bassiana* growth when incubated. No confirmed infection by *Metarhizium* spp. was observed on the insects. Infections by *Entomophaga* also were commonly observed after incubation in cages (Torres-Acosta et al. 2010). The consensus on identification of cryptic species of *Beauveria* and *Metarhizium* indicated use of sequencing by DNA markers such as elongation factor 1 (EF-1) and others; an example is molecular identification of generalist species of *Metarhizium* in the PARB clade (*M. pingshaense* Chen & Guo, *M. anisopliae*, *M. robertsii* Bischoff, Rehner & Humber, and *M. brunneum* Petch) (Bischoff et al. 2009). Identity of *M. acridum* is rather clearly delimited by natural host range, growth habit on hosts, and microscopic morphology.

All dead insects examined collected in the “summit disease” position (Figs. 1-2) had *Entomophaga* resting spores in them. Before incubation at high humidity, the insects did not show external mycelial growth typical of *B. bassiana* or other fungi. Across years, upon microscopic examination immediately after collection, a variable fraction of *Entomophaga*-infected grasshoppers was packed with *Entomophaga* resting spores but also with thin, ascomycetous septate mycelium distinctly different from the thick hyphal bodies of *Entomophaga* fungi. When incubated in moist chambers, *B. bassiana* mycelium and spores typically formed on the grasshoppers after 5 days (Fig. 4). On specific dates, *Beauveria* was observed in as many as 30% of melanopline grasshoppers infected with *E. calopteni* and incubated with high humidity in October-November 2007-2010. No infection by *B. bassiana* was observed without *E. calopteni* (in the same individual) in field-collected, dead insects.



Fig. 4. External growth of *Beauveria* cf. *bassiana* on *Entomophaga caloptenii*-infected melanoploline grasshopper.

Few *E. macleodii* hosts were collected, and *B. bassiana* was not detected on them. No *Metarhizium* infection was observed on dead or live insects at Saltillo.

Internal and external sporulation by *Beauveria* was on two large dead melanoploline grasshoppers extensively colonized by *Beauveria*. Internal sporulation covered the inner wall of the body cavity analogous to internal sporulation by *M. acridum* in grasshoppers (see following sections), only in the last case, external growth was nil.

Infections by *M. acridum* were observed on 20 (50%) *S. nitens* adults collected live in October 2008 and maintained in the laboratory. No *M. acridum* infected dozens of field-collected or laboratory-incubated grasshoppers (>90% Melanoplinae) other than *Schistocerca*. Natural host range restricted to Orthoptera, growth pattern on hosts, and macro and microscopic morphology allowed unambiguous identification of *M. acridum*.

Insects infected by *M. acridum* became reddish and rigid (Fig. 5). Some infected individuals died in elevated positions, hanging from the screen on the side of the cage, somewhat resembling "summit disease" caused by *Entomophaga*; others died on the floor of the cage. After breaking the cephalic, thoracic, and abdominal cuticle and examining the interior of insects, internal sporulation of *M. acridum* was lining the walls of the body cavity (Fig. 6). Sporulation inside a dead host is a trait of *M. acridum* (Magalhaes et al. 2000) and considered an adaptation to low humidity in the environment (Morales-Romano et al. 2009). No external growth or sporulation was observed on the insects; however, we did not provide favorable conditions (i.e., placement in moist chambers) to induce external development of fungi commonly observed on colonized hosts. Internal sporulation was intense (Figs. 5-6) at low humidity (<40% relative humidity) in the laboratory where insects were kept before and after death. From the conditions, and because insects were usually retrieved intact as they were dying, horizontal transmission in cages was unlikely. No cultures of fungi could be isolated; possibly because of their large size, the insect hosts decayed rapidly in the refrigerator before they could be processed.



Fig. 5. Hole (arrow) opened by authors on cuticle of *Schistocerca nitens* adult to show internal sporulation of *Metarhizium acridum* in thorax. No external fungal growth is present. Notice discoloration and pinkish tones on legs, head, and thorax.

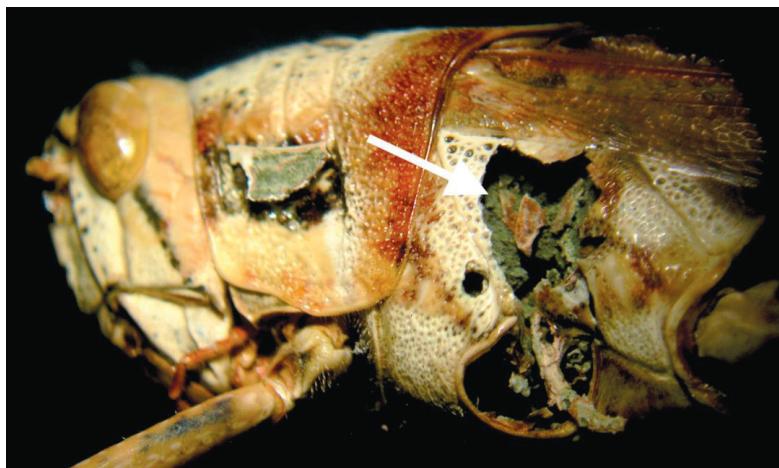


Fig. 6. Extensive internal sporulation of *Metarhizium acridum* visible as green masses (arrow) after breaking thoracic cuticle of *Schistocerca nitens*.

Fungal growth of *M. acridum* in the body cavity of infected *S. nitens* formed a defined fertile layer covering chitinous structures inside the body. The layer consisted of compact, ramified conidiophores. With the aid of a compound microscope (Fig. 7) phialides were cylindrical, 3–4 µm wide, 12–16 µm long ($n = 10$), similar in shape to those in the PARB clade of *Metarhizium* (Bischoff et al. 2009) and to those of the classic concept of *M. anisopliae* sensu lato. Some phialides had a well-defined neck.

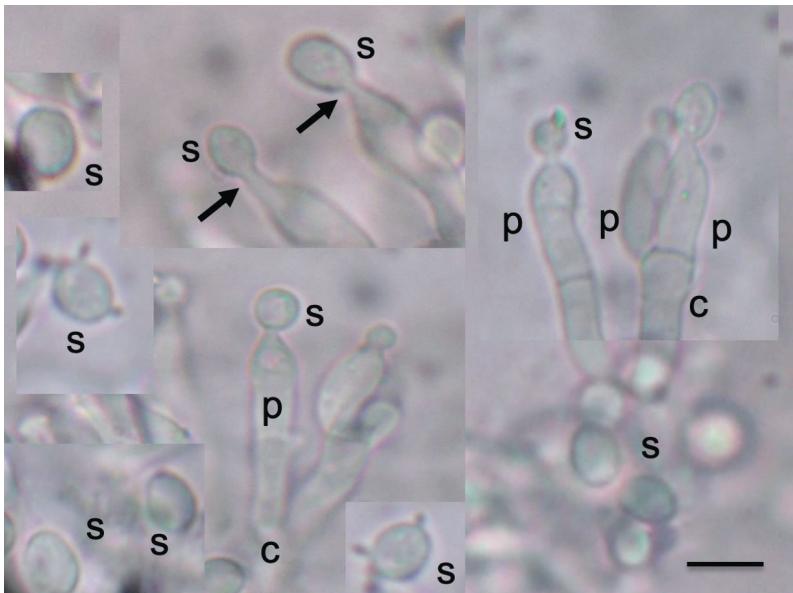


Fig. 7. *Metarhizium acridum* from *Schistocerca nitens*, Saltillo, Mexico: c, branched conidiophores; p, phialides (conidiogenous cells); s, subglobose-ovoid spores (conidia). Spores attached to phialide are not completely developed and are smaller than unattached, mature spores. Arrows = neck on phialides. Bar = 8 μ m.

Conidia were subglobose-ovoid, short-elliptical, 4.0-5.5 \times 3.0-4.0 μ m ($n = 20$), distinctly different from the short-cylindrical conidia of the PARB clade, and also distinct from the short-cylindrical conidia of most *M. acridum* isolates described in Bischoff et al (2009), but similar to those of the type of the species, an African isolate of *M. acridum* (ARSEF 7486 from Benin). Some conidia resembled almost spherical conidia of *Metarhizium globosum*, considered a sister species of *M. acridum*.

One dead *Entomophaga*-infected nymph of *Schistocerca* was observed in October 2008 clasping a grass stem ("summit disease") at the University fields. It had external conidiation, similar to *Entomophaga grylli* pathotype I (= *E. macleodii*). Conidial and conidiophore morphology suggested infection by the species. Definitive identification of fungi uses DNA markers (Casique-Valdés et al. 2012).

Metarhizium anisopliae sensu lato (i.e., a member of PARB complex) (Bischoff et al. 2009) was observed on one grasshopper (*Melanoplus* sp.) collected dead on 15 October 2008 at the base of a bunch of buffelgrass at a Chihuahuan Desert site of Garcia, NL, at 25.698, -100.631. The grasshopper was reddish (a trait shared with *M. acridum*-infected locusts in the study) but had no external fungal growth nor internal sporulation when collected. External sporulation was observed only when the insect was incubated with humidity in the laboratory (Fig. 8). The fungus produced short-cylindrical spores on large and small patches on the insect cuticle. The host, conidial morphology, and sporulation mode were consistent with the classic concept of *M. anisopliae* and members of PARB clade, and different from *M. acridum* (Bischoff et al. 2009). No infection by *M. anisopliae* was found after observation of hundreds of live and dead grasshoppers collected at Saltillo during 4 years.

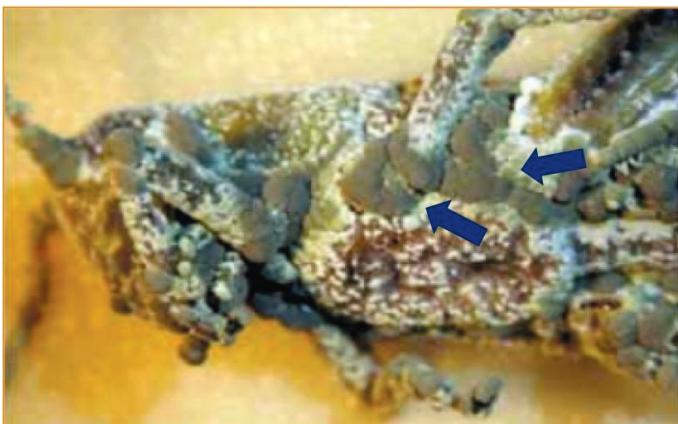


Fig. 8. *Metarhizium* cf. *anisopliae* growing externally (in moist chamber) on *Melanoplus* sp. grasshopper, García, NL, Mexico, October 2008. External production of spores (conidia) on large (arrows) and small patches; compare to internal sporulation in body of *Metarhizium acridum*.

Entomophaga-infected grasshoppers (*Melanoplus* spp.) were observed on 15 November 2021 at Pozo del Gavilán, Galeana, Nuevo Leon (24°48'58.6"N 100°08'01.7"W), 1,600 m above sea level; about 70 km from Saltillo; vegetation was steppe with interspersed pinyon pine, *Pinus cembroides*; and also in Galeana, on 30 October 2021 near La Hedionda, Coahuila, 25°02'57.8"N 100°51'25.4"W, 1,900 m above sea level, 25 km from Saltillo; vegetation was temperate steppe with creosote bush (*Larrea tridentata*), wolfberry (*Lycium berlandieri*), and grasses. Both sites were wide, gently sloped intermontane valleys of the Sierra Madre Oriental. The records suggested wide distribution of fungus in the valleys on arid northern and eastern slopes of the northern Sierra Madre Oriental.

Discussion

In the study, Zoopagomycota (*Entomophaga* spp.) and Ascomycota (*Beauveria* and *Metarhizium* spp.) fungi were found on locusts and other grasshoppers near Saltillo, northeastern Mexico. Insects infected with *Beauveria* and *Metarhizium* spp. often are reddish or pinkish when they die and before external sporulation occurs (Jaronski 2013). In *Beauveria*, it is caused by oosporein antibiotic (Fan et al. 2017). Similar pigmentation was observed in this study (Figs. 4-6).

Biological interaction between *Entomophaga* and *Beauveria* in mixed infections observed is unknown. Both fungi can be primary pathogens of insects. However, there are reports of *Beauveria* strains acting as saprophytic or secondary invaders on insects killed by fungal primary pathogens (Kanga et al. 2004). This is probably the case for *Beauveria* on *Entomophaga* infections reported here.

No *M. anisopliae* sensu lato were found from observation of hundreds of live and dead melanopline grasshoppers collected at Saltillo. Thus, infection by *M. anisopliae* (i.e., PARB complex) seemed very unusual. In the single melanopline grasshopper infected by *M. anisopliae* from Garcia, NL, external sporulation was observed only when the insect was incubated with humidity in the laboratory (Fig. 8).

Extensive collections and sequencing of generalist *Metarhizium* from similar high Chihuahuan Desert locations at Saltillo, near the Garcia, NL site, detected only *Metarhizium robertsii* Bischoff, Rehner & Humber (Guizar-Guzman et al. 2013).

Few, if any, previous images are available specifically of unique internal sporulation of *M. acridum* (Figs. 5-6). The fungus has pantropical distribution: Africa (Kooyman et al 1997), Australia (Hernandez-Velasquez et al. 1997), Brazil (Magalhaes et al. 2000), and Mexican tropics: oceanic Revillagigedo islands and Colima State (Hernandez-Velazquez et al. 1997, Barrientos-Lozano et al. 2002, Pacheco-Hernández et al. 2019), and other sites worldwide.

Internal development and sporulation of *M. acridum* inside grasshopper hosts is adaptation to low humidity (Kooyman 1997, Magalhaes et al. 2000) such as prevalent at the Chihuahuan Desert where we collected. Development inside hosts allows production of myriad fungal spores irrespective of humidity in the environment (Jaronski 2013). When cadavers disintegrate by weather or scavengers, spores are dispersed and infect new hosts.

The known natural host range of *M. acridum* is restricted to Orthoptera (Jaronski 2013); the ARSEF Collection of Entomopathogenic Fungal Cultures catalog (USDA-ARS 2016) lists hosts in Acrididae (Cyrthacanthacridinae, Oedipodinae, and Catantopinae) and Pyrgomorphidae. Melanoplinae seem to be absent as natural hosts. However, individual isolates of *M. acridum* can infect hosts from various orthopteran families different from the original host of the isolates (Magalhaes et al. 2000; Jaronski 2013). This is the first report of the fungus from the Nearctic region shared uninterruptedly by Canada, the United States, and Mexico. The countries include common large, continuous biomes like deserts, prairies, and mountains where grasshoppers cause considerable economic damage to grasslands and crops. Isolates of *M. acridum* demonstrated usefulness in reducing economic infestations by grasshoppers elsewhere (Kooyman et al. 1997, Kamga et al. 2022). However, importation of exotic biological control agents is limited because of concern about possible undesirable impacts on native organisms (Lockwood 1993, Jaronski 2013). Strains of *M. acridum* from *S. nitens* are native to the North American (Chihuahuan) desert biome, and should be exempt from some restrictions, considering lack of natural barriers in biomes across North America. Surveys should be used to detect and isolate native North American strains of *M. acridum*, especially on native *Schistocerca* spp.

Squitier and Capinera (2021) reported fungal pathogens (*Entomophaga* sp., reported as *E. grylli*) attacking only North American species *Schistocerca americana* in Florida. The authors mentioned the fungi can exert significant control of *S. americana* locally. They showed an image of a *S. americana* nymph infected by *Entomophaga* similar to our observation from Saltillo. Squitier and Capinera (2021) also asserted the Florida pathogen did not produce spores externally, a trait of *E. calopteni*. We believe the photograph in Squitier and Capinera (2021) has external, dried mycelium (likely conidiophores) emerging from the abdomen of the nymph, as observed in infection by *E. macleodii*. The known host range of the fungal species is restricted mostly to Oedipodinae (banded-winged grasshoppers). However, its host range might be wider than suspected. We found *E. macleodii* on non-oedipodine hosts (Casique-Valdes et al. 2012); Soper et al. (1982) cited by Carruthers et al. (1997) reported *E. macleodii* was pathogenic to Mesoamerican swarming locust, *Schistocerca piceifrons*. Distribution and ecology of entomopathogenic fungi and grasshoppers at zones of northern Mexico should be studied in detail to support deployment for biological control of orthopteran pests.

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References Cited

- Barrientos-Lozano, L., V. M. Hernández-Velázquez, R. J. Milner, and D. M. Hunter. 2002. Advances in biological control of locusts and grasshoppers in Mexico. *J. Orthoptera Res.* 11: 77-82.
- Bischoff, J. F., S. A. Rehner, and R. A. Humber. 2009. A multilocus phylogeny of the *Metarhizium anisopliae* lineage. *Mycologia* 101: 512-530.
- Casique-Valdes, R., S. R. Sanchez-Peña, R. I. Torres-Acosta, and M. J. Bidochka. 2012. A PCR-based method to identify *Entomophaga* spp. infections in North American grasshoppers. *J. Invertebr. Pathol.* 109: 169-171.
- Fan, Y., X. Liu, N. O. Keyhani, G. Tang, Y. Pei, W. Zhang, and S. Tong. 2017. Regulatory cascade and biological activity of *Beauveria bassiana* oosporein that limits bacterial growth after host death. *Proc. Nat. Acad. Sci. USA* 114: E1578-E1586. doi: 10.1073/pnas.1616543114
- García-Pereyra, J., G. N. Aviña-Martínez, A. A. Orozco-Flores, O. G. Alvarado-Gómez, M. García-Montelongo, G. Alejandre-Iturbide, J. N. Uribe-Soto, and H. Medrano-Roldán. 2014. Variabilidad de cepas de *Metarhizium anisopliae* var. *anisopliae* en base a polimorfismos de ADN amplificados al azar. *Phyton* (Buenos Aires) 83: 37-43.
- Guizar-Guzmán, L., I. Padilla-Guerrero, R. A. Humber, S. R. Sanchez-Peña, and M. Bidochka. 2013. Distribution of the *Metarhizium anisopliae* species complex in Mexico: analysis of Chihuahuan desert, subtropical, and Mesoamerican isolates. In *ESA 61st National Annual Meeting*, Entomological Society of America (D0190), Austin, TX.
- Hernandez-Velazquez, V. M., A. M. Berlanga-Padilla, and E. Garza-Gonzalez. 1997. Detección de *Metarhizium flavoviride* sobre *Schistocerca piceifrons* (Acrididae) en la Isla Socorro, Archipiélago de Revillagigedo, México. *Vedalia* 4: 45-46.
- Jaronski, S. T. 2013. Mycosis inhibits cannibalism by *Melanoplus sanguinipes*, *M. differentialis*, *Schistocerca americana*, and *Anabrus simplex*. *J. Insect Sci.* 13(1): 122. doi: <https://doi.org/10.1673/031.013.12201>
- Kamga, S. F., F. T. Ndjomatchoua, R. A. Guimapi, I., Klingen, C. Tchawoua, A. G. R. Hjelkrem, K. H. Thunes, and F. M. Kakmeni. 2022. The effect of climate variability in the efficacy of the entomopathogenic fungus *Metarhizium acridum* against the desert locust *Schistocerca gregaria*. *Nature Sci. Rep.* 12: 1-15. doi: <https://doi.org/10.1038/s41598-022-11424-0>
- Kanga, L. B., W. A. Jones, R. A. Humber, and D. W. Boyd. 2004. Fungal pathogens of the glassy-winged sharpshooter *Homalodisca coagulata* (Homoptera: Cicadellidae). *Fla. Entomol.* 87: 225-228.
- Kooyman, C., R. P. Bateman, J. Langewald, C. J. Lomer, Z. Ouambama, and M. B. Thomas. 1997. Operational-scale application of entomopathogenic fungi for control of Sahelian grasshoppers. *Proc. Royal Soc. B.* 264: 541-546. doi:10.1098/rspb.1997.0077

- Lockwood, J. A. 1993. Benefits and costs of controlling rangeland grasshoppers (Orthoptera: Acrididae) with exotic organisms: search for a null hypothesis and regulatory compromise. Environ. Entomol. 22(5): 904-914.
- Magalhaes, B., M. Goettel, and E. Da Silva-Frazao. 2000. Sporulation of *Metarhizium anisopliae* var. *acridum* and *Beauveria bassiana* on *Rhammatocerus schistocercoides* under humid and dry conditions. Brazil J. Microbiol. 31: 162-164.
- Pacheco-Hernandez, M. L., J. Resendiz-Martínez, J. Francisco, and V. J. Arriola-Padilla. 2019. Organismos entomopatógenos como control biológico en los sectores agropecuario y forestal de México: una revisión. Revista Mexicana de Ciencias Forestales, 10(56), 4-32. Epub 30 de abril de 2020.<https://doi.org/10.29298/rmcf.v10i56.496>
- Morales-Romano, M. D., C. G. Barajas-Ontiveros, E. M. Del Pozo-Nuñez, M. de Lourdes Rodríguez-Aguilar, and J. J. Nuñez-Lopez. 2009. Condiciones para el desarrollo de *Beauveria bassiana* y *Metarhizium anisopliae* para el control biológico de chapulín frijolero. Tecnociencia Chihuahua 3: 33-38.
- Prior, C., and D. J. Greathead. 1989. Biological control of locusts: the potential for the exploitation of pathogens. FAO Plant Prot. Bull. 37: 37-48.
- Ramoska, W. A., A. E. Hajek, M. E. Ramos, and R. S. Soper. 1988. Infection of grasshoppers (Orthoptera: Acrididae) by members of the *Entomophaga grylli* species complex (Zygomycetes: Entomophthorales). J. Invertebr. Pathol. 52: 309-313.
- Salas-Araiza, M. D, E. Salazar-Solís, and G. Montesinos-Silva. 2003. Acridoideos (Insecta: Orthoptera) del estado de Guanajuato, México. Acta Zool. Mex. 89: 29-38.
- Sanchez-Peña, S. R. 2000. Entomopathogens from two Chihuahuan Desert localities in Mexico. BioControl 45: 63-78.
- Sanchez-Peña, S. R. 2005. In vitro production of hyphae of the grasshopper pathogen *Entomophaga grylli* (Zygomycota: Entomophthorales): potential for production of conidia. Fla. Entomol. 88: 332-334.
- SENASICA. 2022. "Chapulines de Importancia Económica en México en cultivo de frijol: *Brachystola magna*, *Brachystola mexicana*, *Melanoplus differentialis*, *Sphenarium purascens*, *Taeniopoda eques* y *Boopedon diabolicum*". https://www.gob.mx/cms/uploads/attachment/file/600964/Chapulines_de_importancia_econ_mica_en_M_xico.pdf Accessed 15 May 2022.
- Squitier, J., and J. L. Capinera. 2021. Featured creatures: American grasshopper. University of Florida/IFAS Publication EENY-4. https://entnemdept.ufl.edu/creatures/field/american_grasshopper.htm Accessed 19 august 2022
- Torres-Acosta, R. I., S. R. Sanchez-Peña, and G. Gallegos-Morales. 2010. Artificial production of the grasshopper pathogen *Entomophaga calopteni* (*Entomophaga grylli* pathotype 2) in live hosts. 58th Annual Meeting of the Southwestern Branch, Entomological Society of America, Cancun, Mexico.
- USDA-ARS (U.S. Department of Agriculture, Agricultural Research Service) Biological Integrated Pest Management Research Unit. 2016. ARS Collection of Entomopathogenic Fungal Cultures (ARSEF). <https://doi.org/10.15482/USDA.ADC/1326695> Accessed 27 June 2022.

Evaluation of Twenty Maize Germplasms from Belize, French Guiana, Guyana, and Suriname for Resistance to Leaf-Feeding *Spodoptera frugiperda*¹

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Abstract. Fall armyworm, *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae), is native to the Americas and a recent invasive pest of Africa, Asia, and Oceania. The insect causes economic damage to maize (*Zea mays* L.) by feeding on leaf, tassel, and ear tissue. The pest is resistant to several classes of insecticide and Bt-maize grown in certain geographic areas. Native resistance in maize combined with existing control tactics could help overcome control issues. The objective for the study was to evaluate resistance to leaf-feeding fall armyworm in maize germplasm from four countries in the historical range of the pest. Plants grown in the field and artificially infested at the seven-leaf stage were visually scored for leaf-feeding damage at 7 and 14 days post infestation. Scores for Ames 28786 ('GIN II') maize germplasm were not different than the resistant check, GT-FAWCC(C5). In 2020 and 2021, 14-day scores for NSL 283834 ('Guyana 807') (5.8 ± 0.3 and 4.9 ± 0.3 , respectively) were not different than GT-FAWCC(C5) (5.3 ± 0.2 and 4.8 ± 0.3 , respectively). The germplasm sources are moderately resistant to leaf feeding by fall armyworm and might be useful to breeding programs developing resistant maize. In one of 2 years of testing, NSL 283833, NSL 283890, and PI 553088 germplasms received 14-day scores <6.0, and further testing is required to determine their ability to resist leaf-feeding fall armyworm. All maize germplasms tested are tropical and require breeding to adapt progeny for additional research in temperate areas.

Introduction

The fall armyworm, *Spodoptera frugiperda* (Lepidoptera:Noctuidae), is native to the Americas and was accidentally introduced into West Africa in 2016 (Goergen et al. 2016). It spread rapidly to more than 70 countries of Africa, Asia, and Oceania by 2021 (FAO 2021). A pest of 353 plant species (Montezano et al. 2018), Eschen et al. (2021) estimated annual loss to agriculture from fall armyworm in Africa alone at USD 9.4 billion. Preferring maize (*Zea mays* L.), larvae damage leaf, tassel, and ear tissue (Sparks 1979, Marenco et al. 1992). Maize yield loss to fall armyworm can be significant in all regions the pest resides, for example, yield loss of 40% was attributed to fall armyworm in Nicaragua (Wyckhuys and O'Neil 2006) and farmer surveys in Ghana and Zambia estimated national loss at 45 and 40%, respectively (Day et al. 2017).

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Control with insecticide is difficult because application must occur before larvae move to protected areas to feed and because the pest has developed resistance to several classes of insecticide (Yu et al. 2003). Genetically modified maize producing delta-endotoxins from *Bacillus thuringiensis* Berliner (Bt) provides some control of fall armyworm; however, populations of the insect have become resistant to Bt toxins Cry1Ab (Omoto et al. 2016) and Cry1F (Farias et al. 2014) in Brazil, and Cry1F in Puerto Rico (Storer et al. 2010) and the southeastern United States (Huang et al. 2014). Native sources of maize resistance to fall armyworm would enable breeders to develop stable control strategies for the pest. Native sources of resistance have been discovered in tropical maize and incorporated successfully into breeding programs. For example, inbred lines from Mississippi using maize germplasm from Antigua as the resistant donor have been publicly released (CIMMYT, El Batán, Mexico), including Mp708 (Williams et al. 1990) that was well-characterized (Mohan et al. 2006, Womack et al. 2018), publicly available, and often used as a standard check for studies of resistance to fall armyworm. Inbred lines FAW7061 and FAW7111 were developed by Ni et al. (2000) from GT-FAWCC(C5) (Wiseman et al. 1996) and were as resistant as Mp708. Abel et al. (2018) identified four maize germplasms from the island of St. Croix (U.S. Virgin Islands) and five from Thailand (Abel and Scott 2020) that were moderately resistant to the pest. Because fall armyworm adapted to different control tactics, additional sources of maize resistant to the pest would be beneficial. The objective of the study was to test maize germplasm from four countries in the historical host range of the fall armyworm for resistance against leaf feeding.

Materials and Methods

The terms “maize”, “Belize”, “French Guiana”, “Guyana”, and “Suriname” were used 8 January 2020 to search the USDA-ARS GRIN-Global database, and 20 germplasm accessions (Table 1, entry rows 1-20) had available seed. All 20 are landrace populations. GEMN-0131 is a partial inbred also selected as a susceptible check for the study (Abel et al. 2020), GT-FAWCC(C5) (Widstrom et al. 1993) is a population selected as a resistant check, and Mp708 (Williams et al. 1990) is an inbred line selected as a resistant check. On 12 May 2020 and 5 May 2021, maize entries were planted in single-row plots (4.6 x 1 m, 24 seeds/row) at the Agricultural Engineering/Agronomy and Central Iowa Research Farms (Boone, IA) on Nicollet clay loam soil receiving fertilizer pre-plant based on soil tests and a yield goal of 12.5 tons per hectare of grain. The experimental design was a randomized complete block with genotypes as treatments and four replications blocked to control field variation.

Eggs of fall armyworm were acquired from the Corn Host Plant Resistance Research Unit, USDA-ARS, Mississippi State, MS. The colony are maintained on a wheat germ-based diet, and wild adults were added annually to maintain colony vigor. Fall armyworm eggs until hatched were kept at $25 \pm 0.4^{\circ}\text{C}$, $75 \pm 10\%$ relative humidity, and a photoperiod of 14:10 light:dark hours. After eclosion, neonates were added to sterilized corn-cob grits and calibrated so one inoculator “shot” of corn-cob grits contained 15 ± 3 neonates. One calibrated shot was added to the whorl of each V7-V8 stage maize plant (Benson and Reetz 1985) and another calibrated shot was added the next day for a total of approximately 30 neonates per plant. The infestation technique was fully described by Davis et al. (1996). At 7 and 14 days post infestation, a scale of 0 (no damage) to 9 (extensive damage) was used to visually score plants for leaf-feeding damage by fall armyworm (Davis et al. 1992). At 7 days

Table 1. Fall Armyworm Leaf-Feeding Damage Scores 7 and 14 Days Post Infestation of 20 Maize Genotypes and Three Checks Tested at Ames, IA, 2020 and 2021

Genotype	2020		2021	
	FAW 7 days	FAW 14 days	FAW 7 days	FAW 14 days
Ames 28782	5.8 ± 0.3cde	6.6 ± 0.2abcd	6.8 ± 0.3def	6.5 ± 0.3bcde
Ames 28784	5.0 ± 0.3efg	6.2 ± 0.2cdefg	5.8 ± 0.3ghi	6.5 ± 0.3bcde
Ames 28785	6.7 ± 0.4bc	6.5 ± 0.3bcdcf	7.0 ± 0.3cde	6.7 ± 0.3bcde
Ames 28786	4.5 ± 0.3gh	5.6 ± 0.2gh	5.5 ± 0.3hij	5.5 ± 0.3fgh
Ames 28787	7.0 ± 0.4ab	6.6 ± 0.3abcde	7.0 ± 0.4cde	6.0 ± 0.3efg
Ames 28788	5.5 ± 0.3def	6.4 ± 0.2bcdcf	6.5 ± 0.3defg	6.2 ± 0.3def
NSL 283833	4.8 ± 0.3fg	6.4 ± 0.2bcdcf	5.2 ± 0.3ij	5.4 ± 0.3gh
NSL 283834	5.7 ± 0.4cdef	5.8 ± 0.3efgh	6.1 ± 0.4efghi	4.9 ± 0.3h
NSL 283887 CYMT	6.5 ± 0.3bc	6.9 ± 0.2ab	7.8 ± 0.3abc	7.0 ± 0.3bcd
NSL 283888	6.2 ± 0.3bcd	6.9 ± 0.2abc	6.2 ± 0.3efgh	6.9 ± 0.3bcd
NSL 283889	6.0 ± 0.3cd	5.8 ± 0.2efgh	6.0 ± 0.3fghi	7.0 ± 0.3bc
NSL 283890	5.5 ± 0.3def	6.2 ± 0.2cdefg	5.5 ± 0.3hij	5.2 ± 0.3gh
NSL 283891 CYMT	6.0 ± 0.3cd	6.5 ± 0.2bcdcf	6.2 ± 0.3efgh	7.0 ± 0.3bcd
NSL 6550	5.5 ± 0.3def	6.5 ± 0.2bcdcf	6.8 ± 0.3def	6.6 ± 0.3bcde
PI 511561	5.5 ± 0.3def	6.6 ± 0.2abcde	6.0 ± 0.3fghi	6.4 ± 0.3bcde
PI 553088	6.0 ± 0.3cd	5.8 ± 0.2fgh	6.8 ± 0.3def	6.4 ± 0.3bcde
PI 553089	6.8 ± 0.3bc	6.4 ± 0.2bcdcf	7.2 ± 0.3bcd	6.9 ± 0.3bcd
PI 553090	6.2 ± 0.3bcd	6.6 ± 0.2abcde	8.2 ± 0.3a	8.0 ± 0.3a
PI 553091	6.5 ± 0.3bc	6.6 ± 0.2abcde	7.0 ± 0.3cde	7.1 ± 0.3b
PI 692894	5.8 ± 0.3cde	6.1 ± 0.2defg	7.0 ± 0.3cde	6.3 ± 0.3cde
GEMN-0131	7.8 ± 0.3a	7.2 ± 0.2a	8.0 ± 0.3ab	8.1 ± 0.3a
GT-FAWCC(C5)	3.8 ± 0.3hi	5.3 ± 0.2h	4.6 ± 0.4j	4.8 ± 0.3h
Mp708	3.0 ± 0.3i	4.5 ± 0.2i	3.0 ± 0.3k	2.9 ± 0.3i

Means (\pm SE) followed by the same letter in a column are not significantly different according to LSD $P \leq 0.05$.

post infestation, each row was given an overall score. At 14 days post infestation, the first 10 plants in each row were scored individually and the scores averaged per row for analysis. Data were analyzed using PROC GLIMMIX, and means were separated using LSD at $P \leq 0.05$ (SAS Institute 2011).

Results and Discussion

Scores of leaf-feeding damage by fall armyworm at 7 and 14 days post infestation differed significantly for the 24 maize genotypes in 2020 ($F = 9.14$, df = 3,22, $P < 0.0001$; $F = 5.55$, df = 3,22, $P < 0.0001$, respectively) and also in 2021 ($F = 13.35$, df = 3,22, $P < 0.0001$; $F = 15.95$, df = 3,22, $P < 0.0001$, respectively). Scores for Mp708 were significantly less than any of the other 20 germplasm accessions tested (Table 1). All scores for Ames 28786 ('GIN II') maize germplasm were not different than those for the resistant check, GT-FAWCC(C5). In 2020 and 2021, 14-day scores for NSL 283834 ('Guyana 807') were not different than those for GT-FAWCC(C5). NSL 283834 had low germination and only 12 total plants available for testing during 2 years with four replications per year. NSL 283833, NSL 283890, and PI 553088 each had 1 year where they received a 14-day score <6.0 which was not

significantly different from that of the resistant check GT-FAWCC(C5). Retesting of the three germplasms is necessary to determine if they are moderately resistant to leaf feeding by fall armyworm.

Ames 28786 and NSL 283834 were moderately resistant and might be useful for breeding programs interested in adding beneficial alleles to experimental lines being developed to resist leaf feeding by fall armyworm. For temperate maize breeding programs, Ames 28786 and NSL 283834 are tropical and require breeding with temperate germplasm to adapt progeny for research. Both genotypes originated from Guyana. At the time of this study, only four maize genotypes from Guyana were available from the USDA-ARS National Plant Germplasm System for testing. It might be beneficial to search tropical maize seed banks, e.g., International Maize and Wheat Improvement Center (CIMMYT), for additional Guyana maize genotypes to test for resistance to leaf feeding by fall armyworm.

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References Cited

- Abel, C. A., and M. P. Scott. 2020. Evaluation of 21 Thailand maize germplasms for resistance to leaf feeding *Spodoptera frugiperda* (Lepidoptera: Noctuidae). J. Kan. Entomol. Soc. 93: 97-102. doi: 10.2317/0022-8567-93.1.97.
- Abel, C. A., B. S. Coates, and M. P. Scott. 2018. Evaluation of maize germplasm from Saint Croix for resistance to leaf feeding by fall armyworm. Southwest. Entomol. 44: 99-103.
- Abel, C. A., B. S. Coates, M. Millard, W. P. Williams, and M. P. Scott. 2020. Evaluation of XL370A-derived maize germplasm for resistance to leaf feeding by fall armyworm. Southwest. Entomol. 45: 69-74.
- Benson, G. O., and H. F. Reetz. 1985. Corn plant growth-seedling to tasseling, NCH17, pp. 1-3. In National Corn Handbook. Iowa State University Extension, Ames, IA.
- Davis, F. M., S. S. Ng, and W. P. Williams. 1992. Visual rating scales for screening whorl-stage corn for resistance to fall armyworm. Tech. Bull. 186, Mississippi Agricultural and Forestry Experiment Station.
- Davis, F. M., B. R. Wiseman, W. P. Williams, and N. W. Widstrom. 1996. Insect colony, planting date, and plant growth stage effects on screening maize for leaf-feeding resistance to fall armyworm (Lepidoptera: Noctuidae). Fla. Entomol. 79: 317-328.
- Day, R., P. Abrahams, M. Bateman, T. Beale, V. Clottee, M. Cock, Y. Colmenarez, N. Corniani, R. Early, J. Godwin, J. Gomez, P. G. Moreno, S. T. Murphy, B. Oppong-Mensah, N. Phiri, C. Pratt, S. Silvestri, and A. Arne. 2017. Fall

- armyworm: impacts and implications for Africa. *Outlooks on Pest Management* 28: 196-201.
- Eschen, R., T. Beale, J. M. Bonnin, K. L. Constantine, S. Duah, E. A. Finch, F. Makale, W. Nunda, A. Ogunmodede, C. F. Pratt, E. Thompson, F. Williams, A. Witt, and B. Taylor. 2021. Towards estimating the economic cost of invasive alien species to African crop and livestock production. *CABI Agric. Biosci.* 2: 1-18. <https://doi.org/10.1186/s43170-021-00038-7>
- Farias, J. R., D. A. Andow, R. J. Horikoshi, R. J. Sorgatto, P. Fresia, A. C. Santos, and C. Omoto. 2014. Field-evolved resistance to Cry1F maize by *Spodoptera frugiperda* (Lepidoptera: Noctuidae) in Brazil. *Crop Prot.* 64: 150-158.
- FAO (Food and Agriculture Organization of the United Nations). 2021. Global action for fall armyworm control. <http://www.fao.org/fall-armyworm/monitoring-tools/faw-map/en/> Accessed 16 August 2021.
- Goergen, G., P. L. Kumar, S. B. Sankung, A. Togola, and M. Tamo. 2016. First report of outbreaks of the fall armyworm *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera, Noctuidae), a new alien invasive pest in West and Central Africa. *PLOS One* 11: e0165632. doi:10.1371/journal.pone.0165632.
- Huang, F., J. A. Qureshi, R. L. Meagher Jr., D. D. Reisig, G. P. Head, D. A. Andow, X. Ni, D. Kerns, G. D. Buntin, Y. Niu, F. Yang, and V. Dangal. 2014. Cry1F resistance in fall armyworm *Spodoptera frugiperda*: single gene versus pyramided Bt maize. *PLOS One* 9: e112958. doi:10.1371/journal.pone.0112958
- Marencio, R. J., R. E. Foster, and C. A. Sanchez. 1992. Sweet corn response to fall armyworm (Lepidoptera: Noctuidae) damage during vegetative growth. *J. Econ. Entomol.* 85: 1285-1292.
- Mohan, S., P. W. K. Ma, T. Pechan, E. R. Bassford, W. P. Williams, and D. S. Luthe. 2006. Degradation of the *S. frugiperda* peritrophic matrix by an inducible maize cysteine protease. *J. Insect Physiol.* 52: 21-28.
- Montezano, D. G., A. Specht, D. R. Sosa-Gómez, V. F. Roque-Specht, J. C. Sousa-Silva, S. V. Paula-Moraes, J. A. Peterson, and T. E. 2018. Host plants of *Spodoptera frugiperda* (Lepidoptera: Noctuidae) in the Americas. *Afric. Entomol.* 26: 286-300.
- Ni, X., Y. Chen, B. E. Hibbard, J. P. Wilson, W. P. Williams, G. D. Buntin, J. R. Ruberson, and X. Li. 2000. Foliar resistance to fall armyworm in corn germplasm lines that confer resistance to root- and ear-feeding insects. *Fla. Entomol.* 94: 971-981.
- Omoto, C., O. Bernardi, E. Salmeron, R. J. Sorgatto, P. M. Dourado, A. Crivellari, R. A. Carvalho, A. Willse, S. Martinelli, and G. P. Head. 2016. Field-evolved resistance to Cry1Ab maize by *Spodoptera frugiperda* in Brazil. *Pest Manage. Sci.* 72: 1727-1736.
- SAS Institute. 2011. SAS System for Windows. Release 9.2. SAS Institute, Cary, NC.
- Sparks, A. N. 1979. A review of the biology of the fall armyworm. *Fla. Entomol.* 62: 82-87.
- Storer, N. P., J. M. Babcock, M. Schlenz, T. Meade, G. D. Thompson, J. W. Bing, and R. M. Huckaba. 2010. Discovery and characterization of field resistance to Bt maize: *Spodoptera frugiperda* (Lepidoptera: Noctuidae) in Puerto Rico. *J. Econ. Entomol.* 103: 1031-1038.
- Widstrom, N. W., W. P. Williams, B. R. Wiseman, and F. M. Davis. 1993. Registration of GT-FAWCC(C5) maize germplasm. *Crop Sci.* 33: 1422.

- Williams, W. P., G. L. Windham, and F. M. Davis. 1990. Registration of Mp708 germplasm line of maize. *Crop Sci.* 30: 757.
- Wiseman, B. R., F. M. Davis, W. P. Williams, and N. W. Widstrom. 1996. Resistance of a maize population, GT-FAWCC(C5), to fall armyworm larvae (Lepidoptera: Noctuidae). *Fla. Entomol.* 79: 329-336.
- Womack, E. D., M. L. Warburton, and W. P. Williams. 2018. Mapping of quantitative trait loci for resistance to fall armyworm and southwestern corn borer leaf-feeding damage in maize. *Crop Sci.* 58: 529-539.
- Wyckhuys, K. A. G., and R. J. O'Neil. 2006. Population dynamics of *Spodoptera frugiperda* Smith (Lepidoptera: Noctuidae) and associated arthropod natural enemies in Honduran subsistence maize. *Crop Prot.* 25: 1180-1190.
- Yu, S. J., S. N. Nguyen, and G. E. Abou-Elghar. 2003. Biochemical characteristics of insecticide resistance in the fall armyworm, *Spodoptera frugiperda* (J.E. Smith). *Pesticide Biochem. Physiol* 77: 1-11.

Termite Feeding on Aspen and Pine Stakes on a High Elevation Sagebrush-Steppe Rangeland in Southeastern Idaho

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Abstract. An indigenous subterranean termite, *Reticulitermes tibialis* Banks, fed on quaking aspen (*Populus tremuloides* Michx.) and loblolly pine (*Pinus taeda* L.) stakes in prescribed burn and non-burned field plots on an established sagebrush-steppe rangeland restoration study site on Red Mountain, southeastern Idaho, expanding the known distribution of the 'arid-land subterranean termite' species. Aspen and pine stakes in the plots as part of a 5-year wood-decay study were unexpectedly damaged by termites. Groups of 25, 15-cm-long stakes were placed horizontally on the soil surface, and groups of 25, 20-cm-long stakes were vertically inserted to a depth of 20 cm into the soil. Aspen stakes were more damaged by feeding termites than were pine stakes. Stakes inserted into soil sustained greater termite feeding and decay damage compared with stakes on the soil surface. During the first 4 years of the study, wood mass loss caused by termite feeding on stakes exceeded loss from decay. Losses from termite feeding and microbial decay were similar by the 5th year. Differences in termite feeding damage on aspen stakes were compared with pine stakes in prescribed burn and non-burned plots. Stakes in prescribed burn plots had more mass loss than those in non-burned plots.

Introduction

Sagebrush-steppe rangelands encompass thousands of hectares in several areas across the western United States. These rangelands support diverse plant communities, including several species of sagebrush (*Artemisia* spp. L.) and many grasses and forbs (Blaisdell et al. 1982, Miller and Rose 1999, Johnson 2004). Naturally occurring range fires are commonly associated with sagebrush-steppe ecosystems. Widespread prescribed burning as a vegetation management and restoration technique is used to improve habitat for birds and other wildlife (Fernandes et al. 2003). Whereas fire directly impacts the ecology and health of sagebrush-steppe habitats, there is a paucity of information about foraging and feeding behavior by subterranean termites in these ecosystems. Fire can change the amount of woody material on the soil surface and mass of dead roots, thereby

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altering soil properties and affecting ground-dwelling insects. Changes in wood quantity affect wood-feeding insects such as subterranean termites (Hanula et al. 2012).

A 5-year study on Red Mountain in southeastern Idaho evaluated progressive decay severity on groups of 15-cm-long wooden stakes on the soil surface ('surface stakes') and similar groups of 20-cm-long stakes vertically inserted into the soil ('mineral soil stakes'; top end level with the soil surface) in prescribed burn sagebrush-steppe habitat restoration plots and adjacent non-burned 'check' plots. Subterranean termites were feeding in the plots, damaging quaking aspen (*Populus tremuloides* Michx.) and loblolly pine (*Pinus taeda* L.) stakes being evaluated for microbial decay. Termite activity was not expected at the high-elevation, cold winter climate location (winter soil temperature: -2°C at 1-cm depth). Summer daytime air temperatures averaged 12°C (SNOWTEL 2010). Termite species foraging in the plots were identified and their impact on wooden stakes evaluated. Differential influence of prescribed burn sites compared with non-burned sites on termite occurrence and feeding was evaluated.

Materials and Methods

Study Site. The southeastern Idaho study site was on the southwestern and northeastern slopes of Red Mountain (42° 25' 64.5" N – 111° 06' 37.5" E) on the Targhee-Caribou National Forest, Bear Lake County, ~25 km northeast of Montpelier, ID. Elevations were 2,100 to 2,200 m AMSL, with some plots on a 35° (NE) site aspect and others on a 215° (SW) site aspect with a 6-8% slope. For a habitat-restoration study, two treatments were established in 2004: (1) prescribed burn areas, and (2) non-burned areas. To minimize impact to the soil surface, prescribed burns were conducted during spring.

Before prescribed burns, approximate percentages of scattered, patchy ground-cover consisted of interspersed mixtures of little sagebrush (*Artemisia arbuscular* Nutt.; 35%), mountain big sagebrush (*Artemisia tridentata* Nutt. var. *pauciflora* Winward and Goodrich; 30%), prairie Junegrass (*Koeleria macrantha* [Ledeb.] J. A. Schultes; 20%), bluebunch wheatgrass (*Agropyron spicatum* Pursh.; 20%), basin wild rye (*Elymus cinereus* Scribn. and Merr; 10%), yellow rabbitbrush (*Chrysothamnus viscidiflorus* [Hook.] Nutt.; 5%) and mountain snowberry (*Symphoricarpos oreophilus* A. Gray; 1%). Patchy mixtures of scattered forbs including lesser rushy milkvetch (*Astragalus convallarius* Greene; <5%), lupine (*Lupinus sericeus* var. *asotinensis* [Phillips] Hitchc.; <5%), Jones' fleabane (*Erigeron jonesii* Cronquist; 10%), aster (*Symphyotrichum* spp. Nees; 5%), helianthella (*Helianthus uniflorus* Nutt.; 5%), and common yarrow (*Achillea millefolium* L.; 1%) were growing on the study areas. Several grasses interspersed across the study areas were Idaho fescue (*Festuca idahoensis* Elmer; 25%), bluebunch wheatgrass (*Pseudoroegneria spicata* (Pursh) Á. Löve; 5%), western wheatgrass (*Pascopyrum smithii* [Rydb.] Á. Löve; 5%), Kentucky bluegrass (*Poa pratensis* L.; 3%), and New Mexico feathergrass (*Hesperostipa neomexicana* [Thurber ex J. M. Coulter] Barkworth; 5%) (Blaisdell et al. 1982, Miller and Rose 1999, Johnson 2003, Johnson 2004, Cook et al. 2009, Dumroese et al. 2015)

Soil was fine, mixed Pacific Agricryoll with conglomerate and sandstone as parent material. The coarse fraction was 20-35%, with the fine fraction classified as loam (Soil Survey Staff 2003). Mean annual air temperature was ~1.5°C, with

summer daytime air temperatures averaging 12°C and winter soil temperature -2°C at 1-cm deep. Mean annual precipitation was ~78 cm (SNOWTELE 2010).

Termite Identification. Subterranean termites were collected into labelled glass vials containing 90% EtOH and brought to a laboratory for identification and genetic processing. Termite soldiers were collected from stakes on the surface and in mineral soil in prescribed burn and non-burned plots and identified morphologically using standard taxonomic keys (Banks 1946, Gleason and Koehler 1980, Scheffrahn and Su 1994, Brown et al. 2005). Also, soldiers and workers were molecularly identified to species using polymerase chain reaction (PCR) protocols. Termite identification was validated by DNA sequencing and using BLASTn searches on GENBANK. Smith et al. (2010) described the analytical procedures.

Stake Evaluation. Aspen and pine stakes were originally installed to be an index of surface and below-ground impacts of prescribed burning on soil microbes. A detailed description of stake preparation was provided by Jurgensen et al. (2006). During 2004, 600 wood stakes were installed in three replications in each of two treatments (prescribed burn and non-burned 'check'). In each treatment and replication, 25 (2.5 x 2.5 x 15 cm) stakes of each wood species were placed horizontally on the soil surface, and 25 (2.5 x 2.5 x 20 cm) stakes were inserted vertically into the mineral soil. Two subplots were in each replication and main plot. Each year, five stakes of each wood species from each location were collected from prescribed burn and non-burned plots and evaluated for decay and termite damage.

Using stake dimensions, a visual measurement of depth, width and length of wood loss from termite feeding was used to estimate the percentage of stake cross-section (volume) loss, and categorized as: 0% (none), >0–5% (superficial), >5–25%, >25–50%, >50–75%, and >75%. All stakes were cleaned of soil and debris, then oven-dried at 105°C for 48 hours and weighed. Each field stake oven-dry mass was compared with its pre-installation initial dry mass to determine individual and cumulative mass loss caused mainly by termites (no visible decay loss although some decay may have been hidden), decay only (no termite damage),

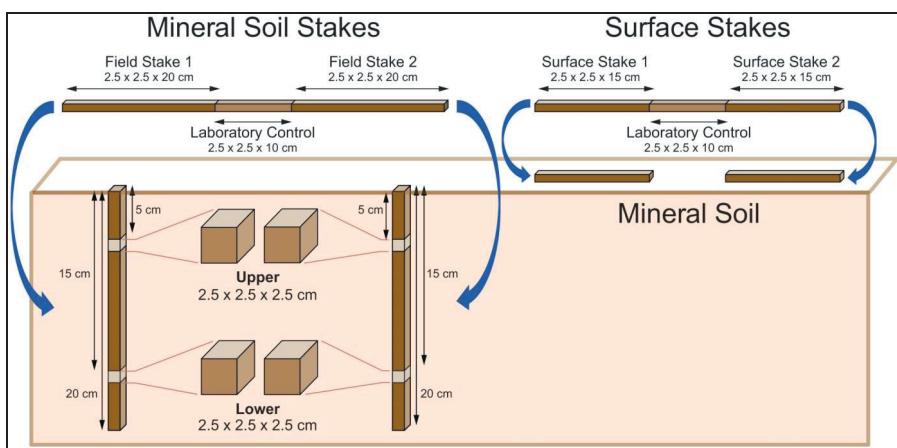


Fig. 1. Stake layout installed horizontally on the soil surface or vertically inserted into mineral soil. Laboratory control sections used for decayed wood and sound wood comparisons (Courtesy Jurgensen et al. 2020).

or obvious decay and termite damage combined for each wood type and field placement (surface or vertically inserted in mineral soil) (Table 1; Figs. 1, 2).

Linear regression was used to evaluate termite feeding on stakes over time and calculate regression formulas (Microsoft 2010). PROC GLM (SAS 2003) was used for analyses of variance for sampling dates (2005-2009), prescribed burn or non-burned, and wood stakes on the surface or in mineral soil.

Results and Discussion

Mass Loss. Subterranean termites fed on aspen and pine stakes on the surface and in mineral soil in prescribed burn and non-burned plots, causing varying amounts of loss of wood mass. Generally, cross-section losses from aspen and pine stakes on the dry soil surface were significantly less compared with stakes in mineral soil ($P \leq 0.01$, Table 1).

When termites were present, mass losses from termite feeding on aspen stakes in mineral soil exceeded loss from decay during the first 4 years, but both termite feeding and decay losses combined were similar by the 5th year, demonstrating that termites are active in recycling dead wood on sagebrush-steppe rangeland (Fig. 2). Overall, aspen stakes were more damaged by feeding and mass loss than were pine stakes ($P \leq 0.01$; Table 1, Fig. 3).

Termite feeding damage was more severe on aspen and pine stakes in mineral soil in prescribed burn compared with non-burned plots. Fewer pine and aspen stakes on the soil surface were damaged by termites compared with those in mineral soil (Table 1). Loss of wood mass of aspen stakes always exceeded that of

Table 1. Number of Stakes Damaged by Termites and Percentage (%) of Stake Cross-section Loss in Prescribed Burn and Non-burned Plots (Study Installed 2004)

Stake and plot type ¹	2005	2006	2007	2008	2009	Total damaged
<u>Pine surface</u>						
Burned	0 (0%)	0 (0%)	1 (<5%)	2 (<5%)	3 (<5%)	6
Non-burned	0 (0)	0 (0)	0 (0)	3 (<5-15)	4 (<5-15)	7
<u>Pine buried</u>						
Burned	1 (<1)	1 (<5)	5 (<5-35)	5 (<5-50)	5 (5-75)	17
Non-burned	1 (<1)	0 (0)	1 (<5)	1 (1-15)	1 (5-15)	4
<u>Aspen surface</u>						
Burned	0 (0)	1(15-25)	2 (35-50)	4 (5->75)	2 (25-75)	9
Non-burned	0 (0)	0 (0)	1 (15-25)	2 (15-75)	3 (25-75)	6
<u>Aspen buried</u>						
Burned	6 (5-35)	7(5-75)	5 (5-75)	6 (15->75)	2 (50->75)	26
Non-burned	0 (0)	1(25-35)	1 (15-25)	0 (0)	2 (5-25)	4

¹Each plot and type had a duplicate group of 25 stakes for each year, with each set remaining in its plot until the year it was collected and evaluated. For example, groups of stakes collected in 2009 were in their plots for 5 years.

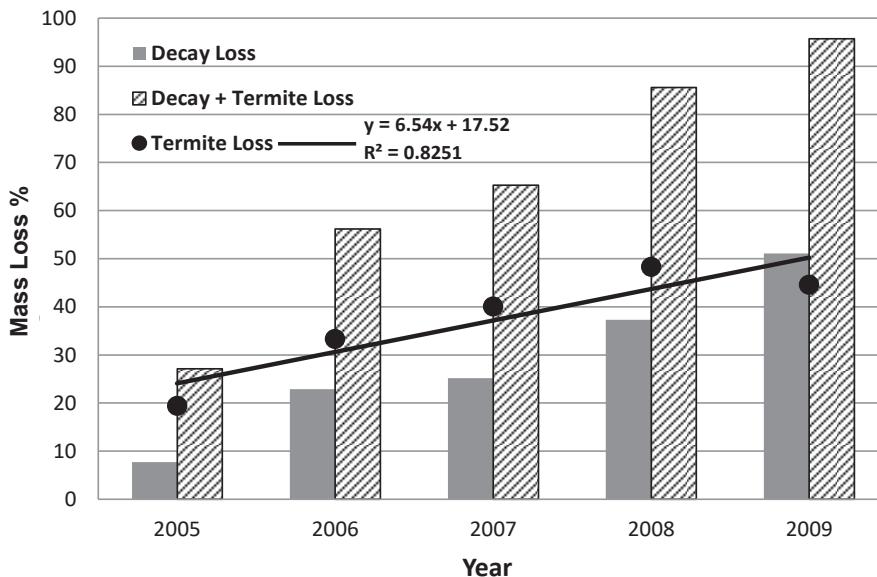


Fig. 2. Increasing percentage of dry mass loss caused by decay, termite feeding, or decay and termite feeding combined on stakes in mineral soil in prescribed burn plots. For example, where termites were feeding on stakes in mineral soil during 2005, an additional 20% mass loss occurred compared with decay alone. The slanted line represents mass loss due to termite feeding.

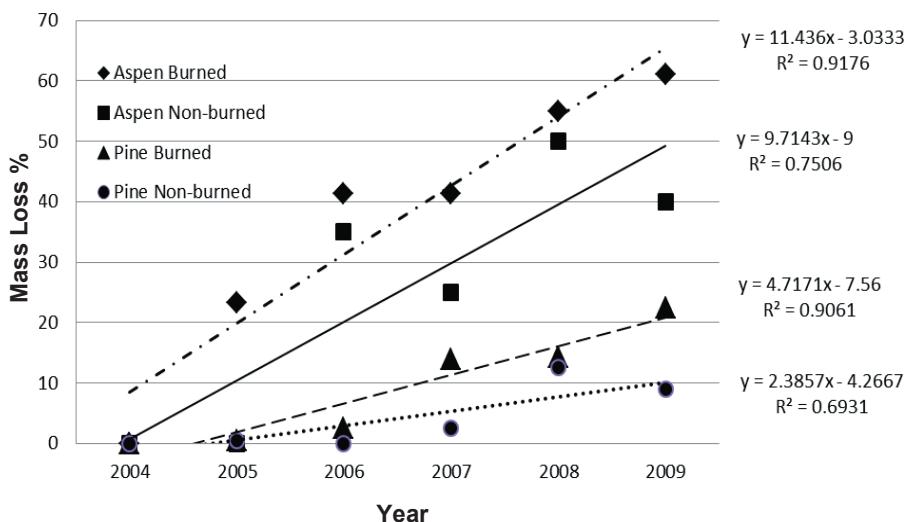


Fig. 3. Average percentage of combined mass loss from termite feeding on aspen and pine stakes on the surface and in mineral soil in prescribed burn and non-burned check plots. Aspen mass loss always exceeded pine mass loss ($P \leq 0.01$).

pine stakes, demonstrating subterranean termites preferred aspen for food. Termites damaged a total of 45 aspen stakes (surface and mineral soil) compared with 34 pine stakes (Table 1, Fig. 3).

During the 5-year study, mean combined mass loss from aspen stakes on the surface and in mineral soil increased from 5 to 72%, greater than the combined mass loss of 2 to 22% from pine stakes on the surface and in mineral soil in prescribed burn and non-burned plots (Fig. 3). For aspen stakes in mineral soil that were damaged by termite feeding and decay, mass loss was approximately double that of decay from microbial activity in mineral soil. Aspen stakes were 20% consumed by termite feeding after 1 year in mineral soil, increasing to 48 and 45% after 4 and 5 years, respectively (Fig. 2). Mean mass loss on the surface increased from 2 to 32% for aspen stakes compared with 1 to 7% for pine stakes.

Stakes in mineral soil were more damaged by termites than were stakes on the soil surface (Table 1). Stakes in mineral soil were exposed to more water, cooler temperatures in summer months and warmer temperatures in winter months. The moderated soil conditions were conducive to termite foraging and microbial proliferation, leading to greater damage to stakes in mineral soil compared with stakes exposed to drier, often hotter or colder more variable microclimate on the surface (Elkins et al. 1986, Nash and Whitford 1995, Li and Su 2008).

When stakes were damaged by termites during the study, average mass loss from damage to aspen always exceeded loss to pine. However, 3 years after installation, numbers of aspen and pine stakes damaged by termites were similar (combined surface and mineral soil) although mass and cross-section losses to aspen continued to exceed those from pine stakes ($P = 0.01$; Table 1, Fig. 3). For example, by the 2nd-year evaluation, only one of five aspen stakes on the surface in prescribed burn plots was damaged by termites, sustaining 15 to 25% cross-section loss, while no pine stakes on the surface were damaged by termites. Contiguous surface stakes with no termite damage had 2.4 to 12% dry mass loss only from microbial decay, demonstrating that after termites locate stakes, damage progression can become severe in a few months (Table 1; Figs. 2, 3).

By the 2nd year, 15 aspen stakes (surface and mineral soil) were damaged by termites in prescribed burn and non-burned plots, while only three pine stakes in mineral soil were damaged by termites. Loss from feeding damage for aspen stakes in mineral soil varied from 5 to >75% of their cross-section. The combined number of aspen stakes on the surface and in mineral soil that were damaged by termites in prescribed burn plots exceeded the number in non-burned plots during the first 4 years, with the trend leveling by the end of the 5th year (Table 1). Cross-section loss for decay alone or decay and termites combined in prescribed burn plots significantly exceeded loss in non-burned plots ($P \leq 0.01$).

By the 5th year, a combined total of 35 aspen stakes on the surface and in mineral soil in prescribed burn plots were more damaged by termites compared with 10 aspen stakes (surface and mineral soil) from non-burned check plots. Termites seemed to forage more actively in prescribed burn plots. Although fewer pine than aspen stakes were damaged by termites, the greatest number of pine stakes damaged by termites were those in mineral soil in prescribed burn plots. During the study, more pine stakes in mineral soil were damaged by termites than were those on the soil surface (Table 1, Fig. 3).

Termites. Termites in pine and aspen stakes were identified morphometrically using a stereo-microscope with computerized measurement software, and also molecular protocols (Banks 1946, Gleason and Koehler 1980,

Scheffrahn and Su 1994, Brown et al. 2005, Smith et al. 2010). On aspen and pine stakes from each location, soldier and worker castes were identified as the 'arid-land subterranean termite' (Snyder 1954, Weesner 1965), *Reticulitermes tibialis* Banks 1920, indigenous to the US Midwest, Great Plains and Northwestern and Pacific Coast states. This is the first record of *R. tibialis* on Red Mountain, ID.

Termite soil excavation and tunneling affect organic matter content and distribution of soil nutrients and porosity (Elkins et al. 1986, Nash and Whitford 1995). Termite tunneling also influences soil bulk density as excavated materials are moved between soil horizons (Li and Su 2008). Redistribution of soil influences plant diversity and viability, and plant biomass and wood accumulation in and on the soil surface, thereby influencing termite foraging (Mando and Stroosnijder 1996, Mando et al. 1999).

Because subterranean termites move vertically between soil horizons, they escape range fires by tunneling downward (Nutting and Jones 1990, Jouquet et al. 2003, Brown et al. 2005, Brown et al. 2009, Morris et al. 2016). Termites contribute to normal background CO₂ and CH₄ flux from soil, affecting soil-dwelling microbe ecology and energy cycles (Konemann et al. 2016). Impacts on sagebrush-steppe rangeland ecosystem soil and plant ecology need additional study.

Subterranean termites inhabiting the Red Mountain study site were adapted to live in high-altitude, sagebrush-steppe rangeland with harsh winter conditions of sub-freezing temperatures and deep snowfall. Results also demonstrated that fire management practices influence occurrence and distribution of subterranean termites in fire-impacted soil, and prescribed burning affected termite foraging and feeding activities in sagebrush-steppe rangeland in southeastern Idaho.

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References Cited

- Banks, F. A. 1946. Species distinction in *Reticulitermes* (Isoptera: Rhinotermitidae). MS thesis, University of Chicago, Chicago, IL.
- Blaisdell, J. P., R. B. Murray, and E. D. McArthur. 1982. Managing intermountain rangelands-sagebrush-grass ranges. USDA For. Serv. Intermountain F abbreviation for intermountain or. Range Exp. Stn.
- Brown, K. S., B. M. Kard, and M. E. Payton. 2005. Comparative morphology of *Reticulitermes* (Isoptera: Rhinotermitidae) of Oklahoma. J. Kan. Entomol. Soc. 78: 277-284.
- Brown, K. S., B. M. Kard, M. E. Payton, and D. R. Kuehl. 2009. Excavation of field-collected soils by *Reticulitermes flavipes* (Isoptera: Rhinotermitidae). J. Kan. Entomol. Soc. 82: 283-292.

- Cook, S. P., F. Merickel, S. M. Birch, and C. Caselton-Lowe. 2009. First report of *Anthicus melancholicus* Laferte (Coleoptera: Anthicidae) in Idaho. Pan-Pac. Entomol. 85: 104-105.
- Dumroese, R. K., T. Luna, B. A. Richardson, F. F. Kilkenny, and J. B. Runyon. 2015. Conserving and restoring habitat for greater sage-grouse and other sagebrush-obligate wildlife: the crucial link of forbs and sagebrush diversity. Native Plants 16: 276-299.
- Elkins, N. Z., T. J. Sabol, T. J. Ward, and W. G. Whitford. 1986. The influence of subterranean termites on the hydrological characteristics of a Chihuahuan Desert ecosystem. Oecologia 58: 521-528.
- Fernandez, P. M., H. Botelho, and S. Nio. 2003. A review of prescribed burning effectiveness in fire hazard reduction. Int'l. J. Wildl. Fire 12: 117-128.
- Gleason, R. W., and P. G. Koehler. 1980. Termites of eastern and southeastern United States: pictorial keys to soldiers and winged reproductives. Bulletin 192. Inst. Food Agric. Sci. (IFAS), Univ. Flor., Gainesville.
- Hanula, J. L., M. D. Ulyshen, and D. D. Wade. 2012. Impacts of prescribed fire frequency on coarse woody debris volume, decomposition, and termite activity in longleaf pine flatwoods of Florida. Forests 3: 317-331.
- Johnson, C. G., Jr. 2004. Alpine and Subalpine Vegetation of the Wallowa, Seven Devils and Blue Mountains. USDA For. Serv. R6-NR-ECOL-TP-03-03.
- Johnson, D. 2003. Red Mountain Prescribed Plan. Bear River Zone Fire Management, Ashton, ID. USDA For. Serv., Caribou-Targhee Nat'l. For.
- Jouquet, P., T. Mery, C. Rouland, and M. Lepage. 2003. Modulated effect of the termite *Ancistrotermes cavithorax* (Isoptera: Macrotermitinae) on soil according to the internal mound structures. Sociobiol. 42: 1-10.
- Jurgensen, M. F., C. A. Miller, and D. S. Page-Dumroese. 2020. Wood decomposition after an aerial application of hydromulch following wildfire in a southern California chaparral shrubland. Front. For. Glob. Change 3: 93.
- Jurgensen, M. F., D. D. Reed, D. S. Page-Dumroese, P. Laks, A. Collins, G. Mroz, and M. Degórski. 2006. Wood strength loss as a measure of decomposition in the northern forest mineral soil. Eur. J. Soil Sci. 42: 23-31.
- Konemann, C. E., B. M. Kard, M. E. Payton, S. P. Deng, J. G. Warren, and T. M. Wilson. 2016. CO₂ emissions from soil on Oklahoma's tallgrass prairie preserve in the presence or absence of termites (Isoptera: Rhinotermitidae). Am. Midl. Nat. 176: 60-71.
- Li, H. F., and N.-Y. Su. 2008. Sand displacement during tunnel excavation by the Formosan subterranean termite (Isoptera: Rhinotermitidae). Ann. Entomol. Soc. Am. 101: 456-459.
- Mando, A., L. Brussaard, and L. Stroosnijder. 1999. Termite- and mulch-mediated rehabilitation of vegetation on crusted soil in West Africa. Restor. Ecol. 7: 33-41.
- Mando, A., and L. Stroosnijder. 1996. Effects of termites on infiltration into crusted soil. Geoderma 74: 107-113.
- Microsoft. 2010. Microsoft Excel. Microsoft, Inc., Redmond, WA.
- Miller, R. F., and J. A. Rose. 1999. Fire history and western juniper encroachment in sagebrush steppe. J. Range Manage. 52: 550-559.
- Morris, C. G., B. M. Kard, R. A. Grantham, A. M. Morris, B. H. Noden, and P. G. Mulder, Jr. 2016. Morphology and foraging behavior of Oklahoma's grass-feeding termite: *Gnathamitermes tubiformans* (Isoptera: Termitidae). J. Kan. Entomol. Soc. 89: 358-372.

- Nash, M. H., and W. G. Whitford. 1995. Subterranean termites: regulators of soil organic matter on the Chihuahuan Desert. *Biol. Fertil. Soils* 19:15-18.
- Nutting, W. L., and S. C. Jones. 1990. Methods for studying the ecology of subterranean termites. *Sociobiol.* 17: 167-189.
- SAS. 2003. Version 2.3. SAS Institute, Inc., Cary, NC.
- Scheffrahn, R. H., and N.-Y. Su. 1994. Keys to soldier and winged adult termites (Isoptera) of Florida. *Fla. Entomol.* 77: 460-474.
- Smith, A. L., M. P. Smith, and B. M. Kard. 2010. Oklahoma Formosan termite surveillance program and termite survey (Isoptera: Rhinotermitidae, Termitidae). *J. Kan. Entomol. Soc.* 83: 248-259.
- SNOTEL. 2010. USDA Nat. Res. Cons. Serv.
<http://www.wcc.nrcs.usda.gov/nwcc/site?sitenum=493&state=id>
- Snyder, T. E. 1954. Order Isoptera: Termites of the United States and Canada. Nat'l. Pest Cont. Assoc., Elizabeth, NJ.
- Soil Survey Staff. 2003. USDA Nat. Res. Cons. Serv.
<http://websoilsurvey.nrcs.usda.gov/>
- Weesner, F. M. 1965. The Termites of the United States: A Handbook. Nat'l. Pest Cont. Assoc., Elizabeth, NJ.

Antioxidative Properties of Ethanolic Extracts Differing by Sex and Coloration of *Pterophylla beltrani*¹

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Abstract. Phenolic compounds are a natural product of plants, many of which are foraged by insects and used for different purposes, e.g., defense against foraging, chitin hardness, antioxidants, pigment production, and antimicrobial synthesis. We extracted metabolite ethanol from *P. beltrani*, quantifying the total phenol content and determining its antioxidative ability by DPPH, ABTS, and FRAP assays. UPLC was used for phytochemical analysis to quantify and characterize major secondary metabolites extracted. The study shows the TPC content varied according to sex ($F_{1,48} = 4.37, P < 0.05$), being significantly more in ethanolic extracts from females. Female-based ethanolic extracts also had greater radical scavenging ability ($F_{2,48} = 3.24, P < 0.05$) than those from males ($F_{1,48} = 9.45, P < 0.05$). The phytochemical profile of the compounds was consistent despite the diet or color of the insect.

Introduction

Through evolution, insects developed diverse adaptive mechanisms for interaction with plants, thus enabling diet selection. Through ingestion of plant-based nutrients or by de novo synthesis, insects obtain secondary metabolites involved in phagostimulation, antioxidant activity, pheromone precursors, photoperiodic-related compounds, UV protection, toxicity, aposematic signals, and coloration (Bernays and Chapman 2000, Heath et al. 2013, Beran et al. 2019). Secondary metabolites include carotenoids, terpenoids, alkaloids, organic acids, cyanide glycosides, sulfate compounds, and phenolic compounds (Opitz and Müller 2009, Beran et al. 2019, Zunjarrao et al. 2020).

Some insects naturally eat a diet rich in secondary metabolites that may include phenolic compounds that could affect general physiology and disease protection in insects (Smilanich et al. 2011, Erler and Moritz 2016, Pusceddu et al. 2018). Most phenolic compounds are of plant origin and might thus be absorbed by

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foraging insects; however, these are often modified into intermediary compounds accumulated in the body of the insect (Nurraihana et al. 2018).

Flavonoids are phenolic compounds naturally produced by plants and an integral part of insect diet and have functions such as defense, chitin hardening, antioxidant activity, and pigment synthesis (Andersen 2010, Musundire et al. 2014, Simonet et al. 2016). The Mexican katydid, *Pterophylla beltrani* (Bolivar and Bolivar), can adapt its diet by changing its nutrition source during the adult stage (Torres-Castillo et al. 2016), an ability that seems linked with transition to the reproductive stage during which it feeds, almost exclusively on three oak species, i.e., *Quercus rysophylla* (Weath), *Q. polymorpha* (Schltdl & Cham), and *Q. canbyi* (Trel).

During the past few years, the insect could be used to develop food supplements and other biotechnologically interesting products because of phenolic compounds and antioxidative agents (Montiel-Aguilar et al. 2017, Rosales-Escobar et al. 2018, García-García et al. 2019, Montiel-Aguilar et al. 2020). However, behavior of the phenolic compounds and secondary metabolites in ethanolic extracts of *P. beltrani* is not fully understood, because their possible role is not known in feeding habits or color of the insect. This study quantified and characterized phenolic compounds, antioxidative agents, and secondary metabolites in ethanolic extracts of *P. beltrani* male and female adults during their change in diet and color, thus contributing to understanding ecological and physiological characteristics.

Materials and Methods

Specimens of *P. beltrani* adults were captured in the northeast zone of Victoria, Tamaulipas; Mexico. The region is known as “La Peregrina” canyon ($23^{\circ}47'13.94''$ N, $99^{\circ}11'45.74''$ O) with vegetation characterized by low subcaducifolious forests, Tamaulipan thorn scrubs, and oak-pine forests (Sánchez-Reyes et al. 2016).

P. beltrani specimens were captured in a *Quercus* spp. forest during three seasons to cover the variable dietary periods of the species (García-García et al. 2019), i.e., early June (spring) for young adults (Coloration 1), July (summer) for adults in transition (Coloration 2), and in autumn for adults (Coloration 3) (Fig. 1). Coloration 1 was characterized by green color dominant throughout the body of the

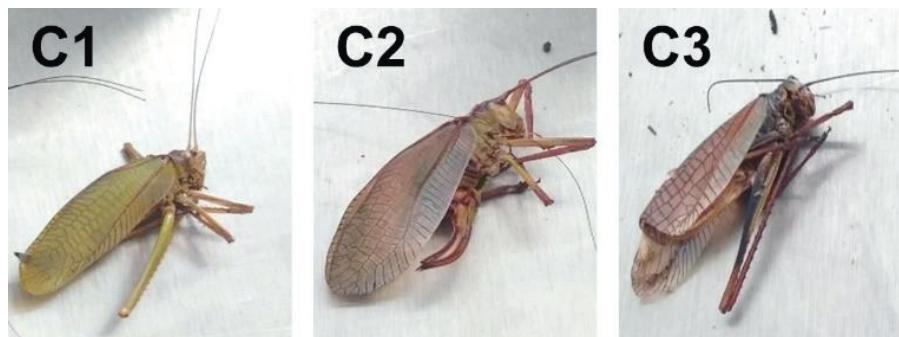


Fig. 1. Types of coloration in *Pterophylla beltrani* (C1, green color dominant throughout the body; C2, reddish or pink wing and legs; C3, reddish body with cherry or pink hues).

insect, Coloration 2 was red or pink legs and wings, while insects with Coloration 3 were reddish with cherry or pink hues. The capture method used in the study was described by Torres-Castillo et al. (2016). Four 100 m² sampling areas were established and specimens collected during 30 minutes. Adults captured were stored in a polyethylene sac and transported live to a laboratory for later processing.

In the laboratory, the specimens were frozen at -20°C for 10 minutes and eviscerated, collecting all the intestinal tissue and nondigested vegetal material to prevent contaminating agents from biasing the analysis (Torres-Castillo et al. 2018). The insects were dried at 45°C for 96 hours. Thirty individuals were put into three groups according to sex and type of coloration. The insects were ground in a mortar and sifted (710 µm/25 mesh sieve, WS Tyler) to obtain fine, homogenous powder for ethanolic extraction. The samples were stored at -20°C until used.

Powdered samples were resuspended in a 1:4 ratio (w/v) in 50% ethanol. The samples were mixed in a vortex for 1 minute, allowed to rest for 10 minutes at room temperature, and put into a sonicator (60 Hz) (Ultrasonic Cleaner UC-10A) at 35°C for 2 minutes. The samples were centrifuged for 7 minutes at 10,000 g/30°C (Torres-Castillo et al. 2018), and the supernatant analyzed for phenolic compounds and antioxidative agents. Total phenol content (TPC) was quantified using 1N Folin-Ciocalteu reagent (Sigma-Aldrich, St. Louis, MO) and 20% Na₂CO₃ (Singleton et al. 1999, Musundire et al. 2014), including a gallic acid standard curve (1-8 µg/ml) (Sigma-Aldrich, St. Louis, MO), to determine TPC per gram of dry weight. Each reaction used 250 µl of standard or sample solution and 125 µl Folin-Ciocalteu reagent, incubating the mixture for 5 minutes at room temperature, followed by addition of 625 µl 20% Na₂CO₃ and incubating at room temperature for 2 hours in the dark. Absorbance of samples/standard was measured at 760 nm in a UV/VIS UV-6000 spectrophotometer (METASH, Shanghai, China). TPC was calculated according to a standard curve; results are shown as milliequivalents of gallic acid per gram of dry weight (mEGA/gDW).

Radical scavenging activity of the extracts was measured by a DPPH assay (2,2-difenil-1-picrylhydrazyl) (Sigma-Aldrich, St. Louis, MO) described by Brand-Williams et al. (1995). Absorbance of 0.02% DPPH methanol solution was adjusted to 0.7 (515 nm) by adding methanol. A standard curve was prepared using Trolox (0.1-1.2 mM) (Sigma-Aldrich, St. Louis MO, EE. UU). The test used 25 µl of sample or standard and 975 µl of DPPH solution, incubating the mixture at room temperature for 30 minutes in the dark. Absorbance of samples or standard was measured at 515 nm, extrapolating into mmol Trolox equivalent per gram of dry weight (mmol TE/gDW).

Radical scavenging activity was evaluated against ABTS (2, 2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) (Sigma-Aldrich, St. Louis, MO), as described by Re et al. (1999). The assay measured ability to scavenge free radicals in a solution. Stock solutions of 7 mM ABTS and 2.45 mM K₂S₂O₈ (Sigma-Aldrich, St. Louis, MO) were mixed in a 2:1 ratio and incubated for 12-16 hours at room temperature, adjusting absorbance to 0.7 ± 0.01 (732 nm) by diluting (ABTS+ solution). For the test, 10 µl of sample or standard were added to 1 ml of ABTS+ solution and incubated for 6 minutes at room temperature. Samples were analyzed at 732 nm in a microplate reader (Bio-Rad Benchmark, Hercules, CA). A Trolox standard curve (0.1-1.2 mM) was compared to determine antioxidative ability of samples. Results were reported as mmol Trolox equivalent per gram of dry weight (mmol TE/gDW).

FRAP antioxidative activity assay was described by Benzie and Strain (1996). Solutions 0.3 mM sodium acetate pH 3.6, 10 mM TPTZ (2,4,6-Tri(2-pyridyl)-s-

triazine) diluted in 40 mM HCl (Sigma-Aldrich, St. Louis, MO), and 20 mM FeCl₃-6H₂O. FRAP solution was prepared with 25 ml H₂O, 900 µl TPTZ, 2.5 ml 20 mM FeCl₃-6H₂O, and 25 ml 0.3 mM sodium acetate, incubated for 30 minutes at 37°C in a water bath. The test used 25 µl of sample or standard in 175 µl of FRAP solution and incubated in the dark for 1 hour at 30°C. A Trolox standard curve (0.1-1.2 mM) was used to determine antioxidative ability of the samples. Samples were analyzed at 595 nm in a Bio-rad Benchmark microplate reader. Results were in mmol Trolox equivalent per gram of dry weight (mmol TE/gDW).

The metabolite profile of the samples was compared by thin-layer chromatography analysis using a method modified from Salazar et al. (2008). Samples were extracted using absolute methanol instead of ethanol, as previously described. During analysis, 30 µl of sample extraction, grouped according to sex and coloration, were put in a 20 x 20 cm plate (UNIPLATE®, Analytech, Newark, DE) and covered with 500 µm of silica gel. The samples were separated in a mobile phase of chloroform, methanol, and water (5:4:1). Chromatogram development was in a glass chamber (30 x 23 x 29 cm) for 20 minutes, dried at 50°C for 3 minutes, put in the dark and sprayed with methanol solution of 2 mg/ml DPPH. After 30 minutes, white spots or bands on a purple background indicated antioxidants confirmed visually.

Analysis and identification of secondary metabolites required injection of 10 µl of *P. beltrani* 50% ethanolic extracts in a UPLC coupled with an ACQUITY QDa mass detector (Waters, MA). Conditions were ACQUITY UPLC® CORTECS® C18 column (1.6 µm, 3.0 x 100 mm); mobile phase A (0.1% formic acid) and B (acetonitrile) in gradient flux (0.5 minute, 10.0% A: 90.0% B: 3.5 minutes, 50.0% A: 50.0% B: 6 minutes, 10.0% A: 90.0% B), total time: 6.0 minutes; caudal: 0.3 ml/minute, injection volume: 2 µl, column temperature: 40°C (Olazarán-Santibáñez et al. 2021). Identities were related by mass comparison with previously reported chemical compounds in 50% ethanolic extracts of *P. beltrani* (Torres-Castillo et al. 2018).

Data from DPPH, ABTS, and FRAP analyses were shown as mean ± SD from duplicate experiments repeated three times. Variables were sex (male or female) and coloration (growth stage), i.e., green young adults (Coloration 1), green/pink adults in transition (Coloration 2), and cherry/pink adults (Coloration 3). Data normality was established with Kolmogorov-Smirnov test. Data were evaluated by two-way ANOVA and Tukey's multiple comparison tests, with 95% confidence. SAS software (SAS Institute, Inc. 2012) was used for the analyses.

Results

Total phenol content and antioxidant ability of ethanolic extracts from *P. beltrani* were tested by DPPH, ABTS, and FRAP. All samples from male and female insects had detectable phenolic compounds and antioxidant ability (Table 1).

The Kolmogorov-Smirnov test showed results had normal distribution. When grouped according to sex, TPC in ethanolic extracts was significantly greater in females than males ($F_{1,48} = 4.37, P < 0.05$). Radical scavenging ability measured by DPPH assay also differed significantly by sex ($F_{2,48} = 3.24, P < 0.05$), with ethanolic extracts from females greater than from males ($F_{1,48} = 9.45, P < 0.05$). However, antioxidant ability was greater in extracts from males than from females ($F_{1,48} = 32.18, P < 0.05$) by ABTS assay ($F_{2,48} = 0.76, P > 0.05$). The FRAP assay showed only one interaction ($F_{2,48} = 5.07, P < 0.05$). Overall, ethanolic extracts from females had greater antioxidant ability, which seemed consistent with more TPC in extracts. ABTS assay showed ethanolic extracts from males had greater antioxidant ability.

Table 1. Total Phenol Content (TPC) and Antioxidative Activity (DPPH, ABTS, and FRAP) of Ethanolic Extracts Differing by Sex of *P. beltrani*

	TPC (mGAE/gDW)	DPPH (mmol TE/gDW)	ABTS (mmol TE/gDW)	FRAP (mmol TE/gDW)
Female (F)	0.20 ± 0.02a	7.71 ± 0.12a	28.6 ± 3.9b	24.9 ± 0.2a
Male (M)	0.19 ± 0.02b	7.57 ± 0.06b	31.4 ± 0.3a	25.3 ± 0.4a

mGAE/gDW: milliequivalents of gallic acid per gram of dry weight; mmol TE/gDW: mmol Trolox equivalent per gram of dry weight. Different letters in a column indicate statistical difference ($p < 0.05$).

When grouping ethanolic extracts from *P. beltrani* according to coloration (C1, C2, and C3), TPC increased with growth stage in male and female specimens. Radical scavenging ability, as shown by DPPH assay, was similar between sexes ($F_{2,48} = 2.75$; $P > 0.05$). However, ABTS assay showed increasing antioxidant ability as color of the insects changed ($F_{2,48} = 68.21$ $P < 0.05$). This was indicated by difference in antioxidant ability in F-C3 extract (32.29 ± 1.36 mmol TE/gDW) when compared with F-C2 (28.85 ± 2.44 mmol TE/gDW) and F-C1 (24.56 ± 1.91 mmol TE/gDW) extracts, the latter of which had least antioxidative activity. The trend was also observed in extracts from males. The FRAP assay confirmed antioxidant ability of extracts depended on the color of specimens ($F_{2,48} = 5.18$ $P < 0.05$), demonstrating ethanolic extract F-C3 (27.06 ± 1.81 mmol TE/gDW) had most radical scavenging potential among females, also observed in extracts from males at the same growth stage, M-C3 (27.25 ± 1.48 mmol TE/gDW). Results suggested difference in TPC measured by ABTS and FRAP was significant and increased as coloration of the insect changed (C1 → C3). The DPPH assay showed greater radical scavenging ability the same way, although antioxidative potential was not different between extracts F-C1 (7.81 ± 0.13 mmol TE/gDW) and F-C2 or F-C3 (Table 2).

Thin layer chromatography (TLC) analysis determined if DPPH profiles of different *P. beltrani* ethanolic extracts differed by sex or coloration (Fig. 2). Although profiles of analyzed samples were similar, radical scavenging ability of extracts from male *P. beltrani* were greater, as evidenced by wider central bands compared to those in extracts from females. The effect might be caused by oversaturation of scavenging compounds in the samples or several intermediate metabolite forms, both of which could be responsible for disparity with previously observed results in which

Table 2. Total Phenol Content (TPC) and Antioxidative Activity (DPPH, ABTS, and FRAP) of Ethanolic Extracts of *P. beltrani* Differing by Color

Coloration	TPC (mEGA/gDW)	DPPH (mmol TE/gDW)	ABTS (mmol TE/gDW)	FRAP (mmol TE/gDW)
F-C1	0.18 ± 0.02cd	7.81 ± 0.13a	24.56 ± 1.91c	23.49 ± 1.93b
F-C2	0.21 ± 0.03abc	7.58 ± 0.13bc	28.85 ± 2.44c	24.21 ± 1.47b
F-C3	0.23 ± 0.03a	7.73 ± 0.12ab	32.29 ± 1.36ab	27.06 ± 1.81a
M-C1	0.18 ± 0.02cd	7.61 ± 0.25abc	28.18 ± 1.96c	23.99 ± 1.86b
M-C2	0.19 ± 0.01bcd	7.60 ± 0.15abc	31.40 ± 1.49b	24.70 ± 2.16ab
M-C3	0.21 ± 0.02ab	7.50 ± 0.14c	34.48 ± 1.43a	27.25 ± 1.48a

mGAE/gDW: milliequivalents of gallic acid per gram of dry weight; mmol TE/gDW: mmol Trolox equivalent per gram of dry weight. Means followed by different letters in a column indicate statistical difference ($p < 0.05$).

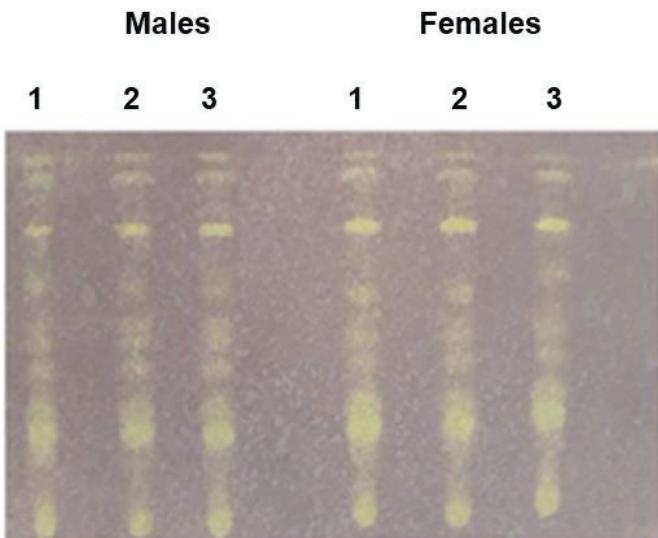


Fig. 2. Radical scavenging ability determined by TLC. Numbers indicate insect coloration. Samples were grouped according to sex. Light bands over dark background indicate compounds with radical scavenging ability.

female extracts had greater antioxidant ability (Table 2). The profile of female-based extracts was more consistent with previous results, showing more bands in F-C3, decreasing gradually and finally displaying fewer bands in F-C1 samples. In comparison, bands in male samples had greater definition, although fewer in number across all types of coloration (Table 1).

Ethanolic extracts from *P. beltrani* also were evaluated by UPLC-MS comparing profiles according to sex and coloration with a *P. beltrani* profile previously obtained in similar conditions (Torres-Castillo et al. 2018). Profile comparison showed no difference between sex or coloration. However, F-C3 extracts had less N-(1-deoxy-1-fructosyl) tryptophan (Table 3).

Discussion

In general, *P. beltrani* has two dietary regimens during its life cycle. The first includes such species as *Pistacia mexicana* (Kunth), *Rhus virens* (Lindh. ex A. Gray), *Brahea berlandieri* (Bartlett), *Chamaedorea radicalis* (Mart), *Sabal mexicana* (Mart), *Flourensia laurifolia* (DC.), *Verbesina persicifolia* (DC.), *Croton niveus* (Jacq), *Caesalpinia mexicana* (A. Gray), *Eysenhardtia polystachya* (Ortega) Sarg, *Havardia pallens* (Benth.) Britton and Rose, *Ocotea tampicensis* (Meissn.) Hemsl, *Fraxinus greggii* (A. Gray), *Colubrina greggii* (S. Watson), *Karwinskia humboldtiana* (Schult) Zucc, and *Ungnadia speciosa* (Endl), while the second includes *Quercus polymorpha* (Schltdl. and Cham), *Quercus canbyi* (Trel), and *Quercus fusiformis* (Small). Dietary change occurs when *P. beltrani* is sexually mature (Torres-Castillo et al. 2018) shown by seasonal changes in color (Fig. 2) and accumulation of phytochemical compounds with unknown ecological or physiological function.

Table 3. Chemical Compounds Detected by UPLC in Ethanolic Extracts of *Pterophylla bettrani* by Sex, Female (F) or Male (M), and Coloration (C1, C2, C3)

Compound	RMW	RT	Coloration					
			F (C1)	F (C2)	F (C3)	M (C1)	M (C2)	M (C3)
Triacanthine	203.11	204	1.0189	+	+	+	+	+
Aminocaproic acid	131.09	130	1.224	+	+	+	+	+
Pantothenic acid	219.11	221	1.113	+	+	+	+	+
2-amino-2-methylbutanoate	117.07	118	1.200	+++	+++	+++	+++	+++
N-(1-deoxy-1-fructosyl) tryptophan	366.14	365	1.1665	+++	+++	+++	+++	+++
2-methoxy-3-(1-methylpropyl) pyrazine	166.11	166	1.2003	+	+	+	+	+
Vasicinol	204.08	-	-	-	-	-	-	-
Alpha-methyl-m-tyrosine	195.08	-	-	-	-	-	-	-
Alpha-cyano-3-hydroxycinnamic acid	189.04	-	-	-	-	-	-	-
Glutamyl-isoleucine	260.13	258	1.141	+++	+++	+++	+++	+++
Acetyl-L-leucine	173.10	175	1.1548	+++	+++	+++	+++	+++
Rhexifoline	207.08	-	-	-	-	-	-	-
Alphitomin	304.05	305	1.1671	+	+	+	+	+
Hericenone A	330.14	-	-	-	-	-	-	-
Perulactone	518.32	518	1.009	+	+	+	+	+
Linoleamide	279.25	279	3.9433	+	+	+	+	+
N-palmitoyl alanine	327.27	-	-	-	-	-	-	-
Sciadonic acid	306.25	307	1.6359	+	+	+	+	+

RMW, reported molecular weight by Torres-Castillo et al. 2018; DMW, detected molecular weight (experimental detection); RT, retention time; F, female; M, male; 1, 2 and 3 indicate the coloration. + (<1 x 10⁵), ++ (>1 x 10⁶), - (negative detection).

Evaluation of TPC and antioxidant ability in ethanolic extracts from *P. beltrani* indicated the content in natural diet. Nondigested vegetal matter was removed before processing samples and contamination discarded, which can be confirmed by comparing with previously observed TPC and antioxidant ability (Torres-Castillo et al. 2016). Other species have potential as a dietary supplement because of antioxidative properties, i.e., *S. piceifrons*, *Acheta domesticus*, *Bombyx mori*, and *Calliptamus italicus* (Di-Mattia et al. 2019, Pérez-Ramírez et al. 2019). The study indicated *P. beltrani* was a source of antioxidants from adult insects regardless of coloration stage.

Results from *P. beltrani* revealed difference in TPC and antioxidant ability, showing ethanolic extracts from females had greater TPC, consistent with previous reports (Di Mattia et al. 2019). Studies of *Polyommatus bellargus* (Rottemburg) (Lepidoptera: Lycaenidae) showed females acquired 67% of the compounds from diet; the compounds are essential in intraspecific visual communication and pigment production. Most of the compounds are stored in posterior wings (to 80%) while it is a small portion of that stored in their bodies (20%). Females of the species sequester greater content than do males regarding visual attraction (Geuder et al. 1997). Because of antibiotic properties, *Polyommatus icarus* (Rott) (Lepidoptera: Lycaenidae) sequesters phenol compounds in the abdomen, head, and thorax, against infection; females of this species also sequester more than males. It was suggested the compounds are accumulated in reproductive organs of the species to provide pigmentation and protection for eggs and later used during eclosion and molting (Schittko et al. 1999).

Accumulation of the compounds in the smaller wings of females might be for visual communication because females with greater content attract more males (Burghardt et al. 2001, Simmonds 2003). This coincides with observations in females of *P. beltrani*, which had more TPC in association with Coloration 3 when insects are in the reproductive phase. Although not proven, this could be associated with diverse functions during mating and oviposition, as observed in other insect species whose reproductive behavior also is affected by phenolic compounds (Atta et al. 2017, Riddick et al. 2018). Overall, TPC in *P. beltrani* was low in comparison with *Henicus whellani* (Chopard) (Orthoptera: Stenopelmatidae) that had TPC of 7.77 mgAE/gDW extracted with water, and *Holotrichia parallela* (Motschulsky) (Scarabaeoidea: Melolonthidae) that had TPC of 33.13 ± 1.49 mgAE/gDW extracted with water (Liu et al. 2012). Although the diet of *P. beltrani* during its adult stage is mostly leaves from various *Quercus* species, many rich in phenolic compounds and tannins, the TPC of the processed specimens do not reflect this equally.

Several biological processes are associated with phenolic compounds and radical scavenging ability, including antioxidative stress mechanisms in some insect species (Dutta et al. 2016, Zielińska et al. 2017). Antioxidative ability of ethanolic extracts from *P. beltrani*, measured by DPPH assay, was higher than in *Schistocerca piceifrons* (Walker) (3.62 ± 0.16 mmol TE/gDW), using the same extraction method (Pérez-Ramírez et al. 2019).

Accumulation of metabolites in Mexican katydid, *Pterophylla beltrani*, is of interest because of dietary adaptation, i.e., diet change to the adult stage. Results of the study showed little variation in content of phenolic compounds in extracts from the species through different growth stages. Females with Coloration 3 had more antioxidant ability in ethanolic extracts, despite change in pigmentation. Concentration of the compounds identified was consistent throughout change in the type of coloration analyzed in the study.

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References Cited

- Andersen, S. O. 2010. Insect cuticular sclerotization: a review. *Insect Biochem. Mol. Biol.* 40: 166-178.
- Atta, E. M., N. H. Mohamed, and A. A. Abdalgawad. 2017. Antioxidants: an overview on the natural and synthetic types. *Eur. Chem. Bull.* 6: 365-375.
- Benzie, I. F., and J. J. Strain. 1996. The ferric reducing ability of plasma (FRAP) as a measure of “antioxidant power”: the FRAP assay. *Anal. Biochem.* 239: 70-76.
- Beran, F., T. G. Köllner, J. Gershenson, and D. Tholl. 2019. Chemical convergence between plants and insects: biosynthetic origins and functions of common secondary metabolites. *New Phytol.* 223: 52-67.
- Bernays, E. A., and R. F. Chapman. 2000. Plant secondary compounds and grasshoppers: beyond plant defenses. *J. Chem. Ecol.* 26: 1773-1794.
- Brand-Williams, W., M. E. Cuvelier, and C. L. W. T. Berset. 1995. Use of a free radical method to evaluate antioxidant activity. *LWT-Food Sci. Technol.* 28: 25-30.
- Burghardt, F., P. Proksch, and K. Fiedler. 2001. Flavonoid sequestration by the common blue butterfly *Polyommatus icarus*: quantitative intraspecific variation in relation to larval hostplant, sex and body size. *Biochem. Syst. Ecol.* 29: 875-889.
- Di Mattia, C., N. Battista, G. Sacchetti, and M. Serafini. 2019. Antioxidant activities in vitro of water and liposoluble extracts obtained by different species of edible insects and invertebrates. *Front. Nutr.* 6: 106.
- Dutta, P., T. Dey, P. Manna, and J. Kalita. 2016. Antioxidant potential of *Vespa affinis* L., a traditional edible insect species of North East India. *PLoS One* 11: e0156107.
- Erler, S., and R. F. Moritz. 2016. Pharmacophagy and pharmacophory: mechanisms of self-medication and disease prevention in the honeybee colony (*Apis mellifera*). *Apidologie* 47: 389-411.
- García-García, L. D., R. I. Torres-Acosta, R. Torres-de los Santos, N. Niño-García, and J. A. Torres-Castillo. 2019. From the forest to the bottle: entomochemicals from *Pterophylla beltrani* (Bolívar and Bolívar) 1 for the beverage Industry. *Southwest. Entomol.* 44: 449-455.
- Geuder, M., V. Wray, K. Fiedler, and P. Proksch. 1997. Sequestration and metabolism of host-plant flavonoids by the lycaenid butterfly *Polyommatus bellargus*. *J. Chem. Ecol.* 23: 1361-1372.
- Heath, J. J., D. F. Cipollini, and J. O. Stireman III. 2013. The role of carotenoids and their derivatives in mediating interactions between insects and their environment. *Arthropod Plant Interact.* 7: 1-20.
- Liu, S., J. Sun, L. Yu, C. Zhang, J. Bi, F. Zhu, and Q. Yang. 2012. Antioxidant activity and phenolic compounds of *Holotrichia parallela* Motschulsky extracts. *Food Chem.* 134: 1885-1891.

- Montiel-Aguilar, L. J., S. R. Sinagawa-García, and J. A. Torres-Castillo. 2017. *Pterophylla beltrani* (Bolívar & Bolívar): prospect for entomophagy. Southwest. Entomol. 42: 591-594.
- Montiel-Aguilar, L. J., J. A. Torres-Castillo, R. Rodríguez-Servin, A. B. López-Flores, V. E. Aguirre-Arzola, G. Méndez-Zamora, and S. R. Sinagawa-García. 2020. Nutraceutical effects of bioactive peptides obtained from *Pterophylla beltrani* (Bolívar & Bolívar) protein isolates. J. Asia-Pac. Entomol. 23: 756-761.
- Musundire, R., C. J. Zvidzai, C. Chidewe, B. K. Samende, and F. A. Manditsera. 2014. Nutrient and anti-nutrient composition of *Henicus whellani* (Orthoptera: Stenopelmatidae), an edible ground cricket, in south-eastern Zimbabwe. Int. J. Trop. Insect Sci. 34: 223-231.
- Nurraihana, H., W. W. Rosli, S. Sabreena, and N. A. Norfarizan-Hanoon. 2018. Optimisation extraction procedure and identification of phenolic compounds from fractional extract of corn silk (*Zea mays* hair) using LC-TOF/MS system. J. Food Meas. Charact. 12: 1852-1862.
- Olazarán-Santibañez, F., G. Rivera, V. Vanoye-Eligio, A. Mora-Olivo, G. Aguirre-Guzmán, M. Ramírez-Cabrera, and E. Arredondo-Espinoza. 2021. Antioxidant and antiproliferative activity of the ethanolic extract of *Equisetum myriochaetum* and molecular docking of its main metabolites (apigenin, kaempferol, and quercetin) on β-tubulin. Molecules 26: 443.
- Opitz, S. E., and C. Müller. 2009. Plant chemistry and insect sequestration. Chemoecology 19: 117.
- Pérez-Ramírez, R., J. A. Torres-Castillo, L. Barrientos-Lozano, P. Almaguer-Sierra, and R. I. Torres-Acosta. 2019. *Schistocerca piceifrons piceifrons* (Orthoptera: Acrididae) as a source of compounds of biotechnological and nutritional interest. J. Insect Sci. 19: 10.
- Pusceddu, M., I. Floris, A. Mura, P. Theodorou, G. Cirotto, G. Piluzza, and A. Satta. 2018. The effects of raw propolis on Varroa-infested honey bee (*Apis mellifera*) workers. Parasitol. Res. 117: 3527-3535.
- Re, R., N. Pellegrini, A. Proteggente, A. Pannala, M. Yang, and C. Rice-Evans. 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic. Biol. Med. 26: 1231-1237.
- Riddick, E. W., Z. Wu, F. J. Eller, and M. A. Berhow. 2018. Utilization of quercetin as an oviposition stimulant by lab-cultured *Coleomegilla maculata* in the presence of conspecifics and a tissue substrate. Insects 9: 77.
- Rosales-Escobar, O. E., B. Villanueva-Bocanegra, J. A. Torres-Castillo, L. U. Arellano-Méndez, and Y. R. Moreno-Ramírez. 2018. Valor antioxidante de la grillete *Pterophylla beltrani* para suplementar alimentos. Southwest. Entomol. 43: 475-483.
- Salazar, R., M. E. Pozos, P. Cordero, J. Perez, M. C. Salinas, and M. Waksman. 2008. Determination of the antioxidant activity of plants from Northeast Mexico. Pharm. Biol. 46: 166-170.
- Sánchez-Reyes, U. J., S. Niño-Maldonado, L. Barrientos-Lozano, and F. M. Sandoval Becerra. 2016. Influencia del clima en la distribución de Chrysomelidae (Coleoptera) en el Cañón de la Peregrina, Tamaulipas, México. Entomol. Mex. 3: 467-473.
- SAS Institute, Inc. 2012. Statistical Analysis System. Version 9.4. SAS Institute, Inc., Cary, NC.

- Schittko, U., F. Burghardt, K. Fiedler, V. Wray, and P. Proksch. 1999. Sequestration and distribution of flavonoids in the common blue butterfly *Polyommatus icarus* reared on *Trifolium repens*. *Phytochemistry* 51: 609-614.
- Simmonds, M. S. 2003. Flavonoid-insect interactions: recent advances in our knowledge. *Phytochemistry* 64: 21-30.
- Simonet, P., K. Gaget, N. Parisot, G. Dupont, M. Rey, G. Febvay, and F. Calevro. 2016. Disruption of phenylalanine hydroxylase reduces adult lifespan, and fecundity and impairs embryonic development in parthenogenetic pea aphids. *Sci. Rep.* 6: 34321.
- Singleton, V. L., R. Orthofer, and R. M. Lamuela-Raventos. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Meth. Enzymol.* 299: 152-178.
- Smilanich, A. M., J. Vargas, L. A. Dyer, and M. D. Bowers. 2011. Effects of ingested secondary metabolites on the immune response of a polyphagous caterpillar *Grammia incorrupta*. *J. Chem. Ecol.* 37: 239-245.
- Torres-Castillo, J. A., S. R. Sinagawa-García, G. Ruiz-De-La-Cruz, A. K. Gámez-Huerta, M. C. Juárez-Aragón, M. Lara-Villalón, and A. Mora-Olivio. 2016. Digestive proteinases and antioxidant capacity from *Pterophylla beltrani* Bolívar & Bolívar fed two natural diets. *Southwest. Entomol.* 41: 613-625.
- Torres-Castillo, J. A., S. R. Sinagawa-García, R. I. Torres-Acosta, L. D. García-García, A. G. Ramos-Rodríguez, B. Villanueva-Bocanegra, and Y. R. Moreno-Ramírez. 2018. Entomochemicals from *Pterophylla beltrani* Bolívar and Bolívar: antioxidants and other metabolites. *Southwest. Entomol.* 43: 369-381.
- Zielińska, E., M. Karaś, and A. Jakubczyk. 2017. Antioxidant activity of predigested protein obtained from a range of farmed edible insects. *Int. J. Food Sci.* 52: 306-312.
- Zunjarrao S. S., M. B. Tellis, S. N. Joshi, and R. S. Joshi. 2020. Plant-Insect Interaction: The Saga of Molecular Coevolution. In J. M. Mérillon and K. Ramawat [eds.], *Co-Evolution of Secondary Metabolites*. Reference Series in Phytochemistry. Springer, Cham.

Capture of *Anastrepha striata* Schiner¹ with Pheromones and Organic Insecticides and Guava Phenology

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Abstract. Guava (*Psidium guajava* L.) is an economically important crop in the State of Guerrero. A study was done in a commercial guava orchard with a history of severe damage from fruit fly infestations, in the town of Coaxtlahuacán, municipality of Mochitlán, Guerrero. The objective of the experiment was to evaluate incorporation of recycled plastic artisan traps with chemical attractants for pest management. Treatments were the attractant CeraTrap® and the insecticide Azanim® in the traps. Infestations were evaluated every 15 days from July 2018 to February 2019, with the number of flies captured and phenology of guava plants recorded. Data were subjected to analysis of variance (ANOVA) and Tukey means test ($P \leq 0.05$). Months with most pests were July (960 adult flies) and December (1,057 adult flies); treatments with greatest efficiency as attractants were 100 ml of CeraTrap® and 150 ml of CeraTrap® (18.74 and 21.49 flies per trap, respectively).

Introduction

Guava (*Psidium guajava* L.) is part of a group of tropical fruit most produced worldwide (Altendorf 2017). Mexico is fifth in production (SADER 2019a). Production in 2015 was 224,841 tons valued at 14,694,000 dollars in exports to Canada, the United States, Guatemala, and Russia, among others (SADER 2017). In Mexico, in 2019 the main guava-producing states were Michoacán, Aguascalientes, and Zacatecas; Guerrero was sixth (SADER 2019b).

In the State of Guerrero, the crop dominates the North zone. Management limitations are phytosanitary and lack of knowledge of phenological stages of the crop. The fruit fly is the most important pest, causing 70% loss in orchards and backyard trees in all production regions (SADER 2020).

¹Diptera: Tephritidae

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Damage by fruit flies begins when at fertile female oviposits eggs below the fruit rind; when mature, larvae hatch and emerge, feeding until the third instar on pulp, causing immature fruit to drop. Once in the ground, the larva leaves the fruit and buries itself a few centimeters to pupate; 15 to 20 days later an adult emerges to start a new cycle (Segura et al. 1995).

Insecticide is the main method for pest management. Application of malathion is recurrent to combat the pest. More environmentally friendly methods that contribute to production of healthier and safer food are being sought (Arias 2015). One of the most successful and used practices is wet traps (water, attractant, hydrolyzed protein) to capture fruit flies (Barrera and Montoya 2007).

For farmers at Coaxtlahuacán, municipality of Mochitlán, Guerrero, cultivated guava is an important source of income, however, inadequate control of the fruit fly and ignorance of the phenology of the crop lead to poor-quality, unsafe products, in addition to low yields. Many commercial orchards in the region have been abandoned. The objective of this research was to evaluate use of artisan traps with approved attractants and insecticides as management treatments to capture fruit flies in organic crops, as well as to record phenological behavior of the crop and incorporate both into an integrated crop management system.

Materials and Methods

Research was done at Coaxtlahuacán, municipality of Mochitlán, Guerrero, at latitude 17° 23' 37", longitude 99° 20' 50", and an altitude of 1,730 m above sea level. The prevailing climate is temperate subhumid C (w2) (w) (SCT-SEMARNAT 2017).

The study was done from July 2018 to February 2019 in a commercial guava orchard with 200 8-year-old trees, propagated by layering and established in a true frame topological arrangement 6 m between trees. Plastic 600-ml bottles (recycled) were used to make traps; the bottles were drilled above the middle with three 1-cm² holes. Inside the trap bottles, the treatments were deposited with a funnel. The bottles were hung with galvanized wire from the main branch in the middle of the tree canopy, with 15 cm between the bottle and branch. Treatments evaluated were: T1 (100 ml of CeraTrap® + 3 ml of Azanim®), T2 (100 ml of CeraTrap® + 6 ml of Azanim®), T3 (100 ml of CeraTrap® + 9 ml of Azanim®), T4 (100 ml of CeraTrap®), T5 (150 ml of CeraTrap® + 3 ml of Azanim®), T6 (150 ml of CeraTrap® + 6 ml of Azanim®), T7 (150 ml of CeraTrap® + 9 ml of Azanim®), T8 (150 ml of CeraTrap®) and checks T9 (3 ml of Azanim ® + 100 ml of water), T10 (6 ml of Azanim ® + 100 ml of water), T11 (9 ml of Azanim ® + 100 ml of water), and T12 (only 100 ml of water). The treatments were replaced every 15 days (to avoid degradation), and the number of flies captured per trap was recorded. For counting and identification, the flies were deposited in 250-ml flasks containing 100 ml of 70% alcohol.

For phenology, seven trees were selected and evaluated from 15 September 2018, to 16 February 2019. Four branches 1.5 to 2.0 cm at the base and 80 to 100 cm long directed toward the four cardinal points were chosen. Variables evaluated were tender button (light green buttons 2-3 mm long), ripe button (yellowish-green buttons more than 5 mm long, close to anthesis), flower (flower fully open, with white petals, and flower whorls of normal size), fruit set (very small fruit, that already shed the petals), and fruit (developing to physiological maturity and consumption).

To evaluate traditional traps to capture flies, an experimental design of complete random blocks was used, with 12 treatments and six replications. A trap was considered an experimental unit. To record phenological stages of reproductive

behavior of the plant, a randomized complete block design was used, with 12 treatments (sampling dates) and seven replications (trees). A tree was considered an experimental unit. Data obtained in both designs were subjected to analysis of variance and a means comparison test (Tukey, $p \leq 0.05$) with the statistical program SAS 9.3 for Windows.

Flies in 70% alcohol were identified in the Plant Physiology and Biotechnology laboratory of the Faculty of Agricultural and Environmental Sciences of the Autonomous University of Guerrero, with the help of an stereomicroscope (IROSCOPE®, model ES-24) and taxonomic keys by Hernández-Ortiz (1992), Caraballo (2001), and Rodríguez and Arévalo (2015). According to the taxonomic keys used, the main morphological characters of the species are dark brown spots in the pre and post-sutural regions of the shield, and scutellum without spots. Medium tergite with dark spots, S and C bands connected at the level of the R4 + 5 vein, S-band separated from V-band vertex, a proximal arm of V-band not remarkably wide (Hernández-Ortiz 1992). The tip of the aculeus rounded apically, with margins without denticulation (Fig. 1).

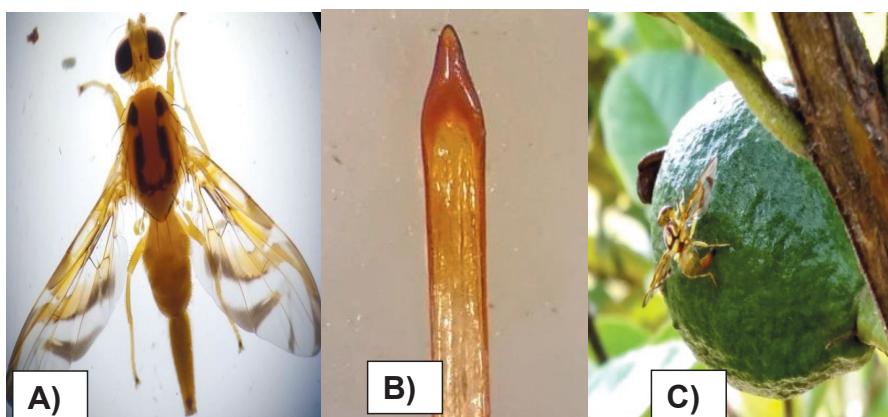


Fig. 1. A) Characteristics of *Anastrepha striata*, dark brown spots in the pre- and post-sutural regions of the scute and scutellum without spots. Medium tergite with dark spots, S and C bands connected at the height of the R4+5 vein. S band separated from apex of V band, proximal arm of V band not noticeably wide. B) Tip of aculeus apically rounded, with margins without denticulation. C) Myrtaceous fly in the study orchard at Coaxtlahuacan, Municipality of Mochitlán, Guerrero.

Results and Discussion

Analysis of variance showed significant differences ($P \leq 0.05$) in the number of fruit and total number of fruit per branch. Most fruits set per branch was on 15 September (2.72 fruits), while January and February had fewest values (0.76 and 1.12 fruit per branch). The greatest number of fruits per sample, was on 27 October (5.79 per branch), while fewest (0.94 and 2.72 fruits per branch) were in January through February (Fig. 2).

Abundance of flies peaked on 21 July (13.33 and 24 November (14.68 flies). Statistically large numbers were on 27 October and 10 November (11.04 and 10.03 flies) and 8 December (10.86). Fewest were on 4 August and 15 September (2 and 3.04 flies) and 22 December and 16 February 2019 (0.1 and 3 flies) (Fig. 3).

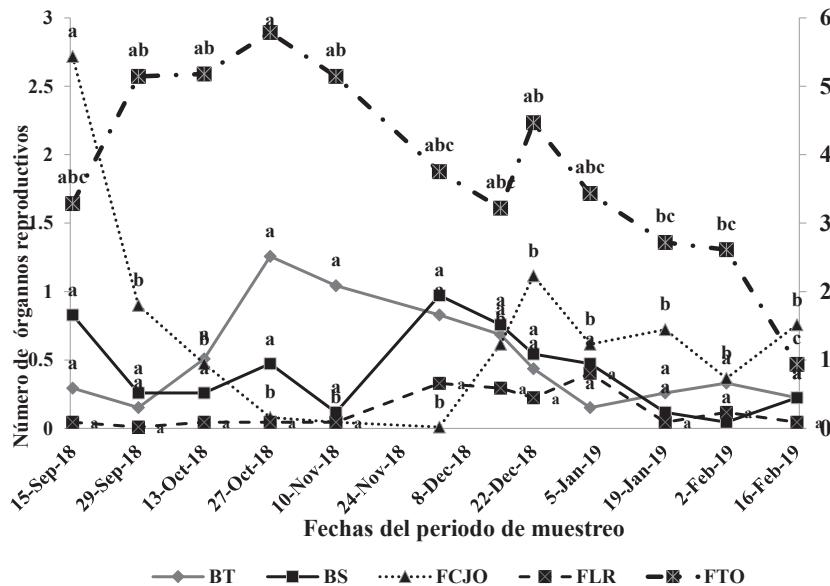


Fig. 2. Phenological stage of reproductive organs of guava. BT (tender buttons), BS (ripe buttons), FCJO (set fruit), FLR (flowers), FTO (harvested fruit). Values with the same letter are not significantly different (Tukey, $P \leq 0.05$).

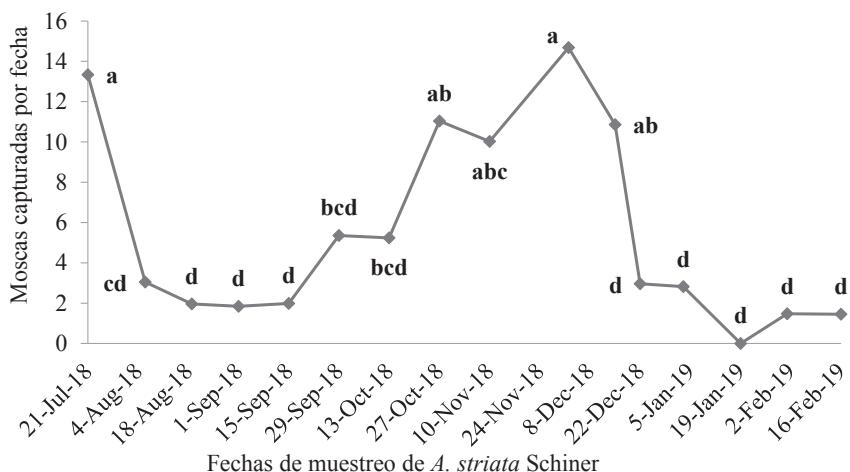
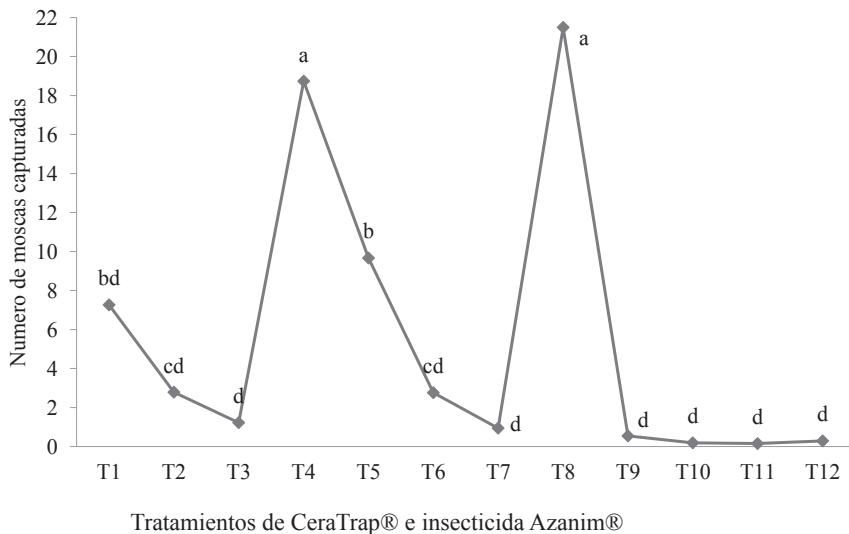


Fig. 3. Abundance of *Anastrepha striata* by sampling date in at guava crop. Values with the same letter are not significantly different (Tukey, $P \leq 0.05$).

Treatments 4 (100 ml) and 8 (150 ml) of CeraTrap® ($P \leq 0.05$) captured the most insects (18.74 and 21.49 flies per trap), were equally effective, and significantly better than other treatments (Fig. 4). T3, T7, T9, T10, T11, and T12 did not significantly affect the numbers of flies captured, and were not recommended for use.



Tratamientos de CeraTrap® e insecticida Azanim®

Fig. 4. Effect of CeraTrap® and Azanim® attractant treatments to trap fruit flies in an organic guava orchard at the town of Coaxtlahuacán, Municipality of Mochitlán, Guerrero, Mexico (Tukey, $P \leq 0.05$). Values with the same letter are not significantly different. T1 (100 ml CeraTrap® + 3 ml Azanim®), T2 (100 ml CeraTrap® + 6 ml Azanim®), T3 (100 ml CeraTrap® + 9 ml Azanim®), T4 (100 ml CeraTrap®), T5 (150 ml CeraTrap® + 3 ml Azanim®), T6 (150 ml CeraTrap® + 6 ml Azanim®), T7 (150 ml CeraTrap® + 9 ml Azanim®), T8 (150 ml CeraTrap®), T9 (3 ml Azanim® + 100 ml water), T10 (6 ml Azanim® + 100 ml water), T11(9 ml Azanim® + 100 water) and T12 (100 ml water).

Discussion

At the town of Coaxtlahuacán, floral organs of guava began to form November 2018 with the peak of flowering in the first days of January (0.4021 flowers per branch). Something similar occurred in cultivated guava at Iguala, Gro. where most floral organs formed in winter (December to February) (Damián-Nava et al. 2004). At Coaxtlahuacán, two fruit production periods were, in December (4.46 fruits per branch) and most fruit was produced from September to November (5.14 to 5.78 per branch). At Iguala, with dry tropical environmental conditions, fruit is harvested in May and June, months in which 1.8 fruit is obtained per shoot.

The guava fly, *Anastrepha striata*, has an altitudinal distribution of 300 to 2,200 m (Castañeda et al. 2010). Coaxtlahuacán is at 1730 m, so the species is in an environment conducive to its development and multiplication. At the research site were other favorable conditions that help in biological development of the pest, such

as high humidity, 18 to 30°C temperature, 8 hours of light, and available hosts (Portilla 1994). During the study flies were most abundant from September to December, which coincided with the phenological stage of growth and maturation of guava fruit. However, the fly was present throughout the year, because of appropriate environmental conditions and multiple host species (Vilatuña et al. 2016). The results are consistent with records at other production regions with similar environmental conditions; for example, Aguilar et al. (2011) reported first attacks by fruit flies of fruit were in the last growth phase when yellow portions (seasoning) were present. The behavior documented in this study was consistent with that in regions of Michoacán, Mexico, where flies always were present, with fluctuations throughout the year and greatest abundance from August to December (Miranda-Salcedo et al. 2012).

In this research, the organic attractant CeraTrap® without organic insecticide was effective in capturing by trapping fruit flies and surpassed the Azanim® organic insecticide treatment. The results were similar to those by Santos-Ramos et al. (2012) who found CeraTrap® food attractant captured significant numbers of fruit flies (especially females) and is an efficient alternative to reduce abundance in the crop. It is important to mention that with flies, affected by exposure to natural enemies such as parasitoid wasps of larvae (Braconidae primary parasitoids of immature stages of mainly Lepidoptera, Coleoptera, and Diptera), predators such as ants, birds, and reptiles, and fungal entomopathogens, the trapping strategy is ecologically friendly (Rodríguez and Arévalo 2015).

At Coaxtlahuacán, municipality of Mochitlán, Guerrero, cultivated guava fruited throughout the year, but was most intense in September and October. For most capture of fruit flies in traps, it is sufficient to use CeraTrap® attractant in doses of 100 to 150 ml. Although flies were most abundant in July and December, they were present from July to February, so trapping should be done throughout this period.

References Cited

- Aguilar, S. G., R. R. Granado, y A. J. L. Domínguez. 2011. Uso de los recursos naturales y productivos en el cultivo de la guayaba en la región de Juchipila, Zacatecas. Revista Geográfica de América Central 2: 1-21. <https://www.redalyc.org/pdf/4517/451744820483.pdf>
- Altendorf, S. 2017. Perspectivas mundiales de las principales frutas tropicales, perspectivas, retos y oportunidades a corto plazo en un mercado global pujante. http://www.fao.org/fileadmin/templates/est/COMM_MARKETS_MONITORING/Tropical_Fruits/Documents/Tropical_Fruits_Spanish2017.pdf Consultado 16 Feb. 2020.
- Arias-Hernández, A. 2015. Productos orgánicos en México, CEDRSSA Centro de Estudios para el Desarrollo Rural Sustentable y la Soberanía Alimentaria, palacio de San Lázaro. Consulta Noviembre 2019. <file:///C:/Users/Acer/AppData/Local/Microsoft/Windows/Temporary%20Internet%20Files/Content.IE5/F2DIVW0R/97Productos%20org%C3%A1nicos%20en%20M%C3%A9xico.pdf>
- Caraballo, J. 2001. Diagnosis and pictorial key for the species of the genus *Anastrepha* Schiner, 1868 (Diptera: Tephritidae) of economic importance in Venezuela. Entomotropica antes/formerly Boletín de Entomología Venezolana 16: 157-164. <http://www.bioline.org.br/pdf?em01028>

- Castañeda, M. R., A. F. Osorio, N. A. Canal, and P. E. Galeano. 2010. Species, distribution and hosts of the genus *Anastrepha* Schiner in the department of Tolima, Colombia. *Agronomía Colombiana* 28: 265-271. <https://revistas.unal.edu.co/index.php/agrocol/article/view/18070/18987>
- Damián-Nava, A., V. A. González-Hernández, P. Sánchez-García, C. B. Peña-Valdivia, M. Livera-Muñoz, and T. Brito-Guadarrama. 2004. Growth and phenology of Guava (*Psidium guajava* L.) cv. "Media China" in Iguala, Guerrero. *Revista Fitotecnia Mexicana* 27: 349-358. <https://revistafitotecniamexicana.org/documentos/27-4/7a.pdf>
- Hernández-Ortiz, V. 1992. El género *Anastrepha* Schiner en México (Díptera: Tephritidae) taxonomía, distribución y sus plantas huéspedes. Instituto de Ecología, Sociedad Mexicana de Entomología, Xalapa, México. file:///D:/M%C3%ADa%20tema/guayaba/HernandezOrtiz_et.al_2010_2%20.pdf
- Miranda-Salcedo, M. A., J. Lledo-Fernández, y P. Montoya-Gerardo. 2012. Fluctuación poblacional de moscas de la fruta *Anastrepha* spp. (Diptera: Tephritidae) en el Valle de Apatzingan, Michoacán. <file:///C:/Users/Acer/AppData/Local/Microsoft/Windows/Temporary%20Intern et%20Files/Content.IE5/S3N4OT6M/1004-1008.pdf>
- Portilla, M., G. González, and L. Núñez. 1994. Infestation, survey and identification of fruitflies and their natural enemies in coffee. *Revista Colombiana de Entomología* 20: 261-266.
- Rodríguez, C. P. A., y P. E. Arévalo. 2015. Las moscas de la fruta de importancia económica en Colombia, Boletín técnico Instituto Colombiano Agropecuario, ICA.
- Santos-Ramos, M., A. Bello-Rivera, R. Hernández-Pérez, and D. F. Leal-García. 2012. Effectiveness of bait station MS2® and food attractant CeraTrap® as an alternative for catching fruit flies in Veracruz, México. *Interciencia* 37: 279-283. <https://www.redalyc.org/pdf/339/33922748007.pdf>
- SADER (Secretaría de Agricultura y Desarrollo Rural). 2017. Aumenta 8.2 por ciento de producción de guayaba en México en el último trienio. <https://www.gob.mx/agricultura/prensa/aumenta-8-2-por-ciento-produccion-de-guayaba-en-mexico-en-el-ultimo-trienio> Consultado 3 Enero 2019.
- SADER (Secretaría de Agricultura y Desarrollo Rural). 2019a. Zacatecas, entre los principales estados productores de guayaba. <https://www.gob.mx/agricultura/zacatecas/articulos/zacatecas-entre-los-principales-estados-productores-de-guayaba?idiom=es> Consultado 13 Dic. 2019.
- SADER (Secretaría de Agricultura y Desarrollo Rural). 2019b. Guerrero sexto lugar en producción de guayaba a nivel nacional. <https://www.gob.mx/agricultura/guerrero/articulos/guerrero-sexo-lugar-en-produccion-de-guayaba-a-nivel-nacional?idiom=es> Consultado 28 Oct. 2019.
- SADER (Secretaría de Agricultura y Desarrollo Rural). 2020. Campaña nacional contra moscas de la fruta en el estado de Guerrero. <https://www.gob.mx/agricultura/guerrero/articulos/campana-nacional-contra-moscas-de-la-fruta-en-el-estado-de-guerrero?idiom=es> Consultado 24 Feb. 2020.
- SCT-SEMARNAT. 2007. Modernización del camino Mochitlán – Coaxtlahuacán, tramo: del Km. 0+000 al Km 10+000, pp. 53-73. Manifestación de impacto ambiental.

<http://Sinat.Semarnat.Gob.Mx/Dgiradocs/Documentos/Gro/Estudios/2008/12ge2008v0006.pdf> Consultad 28 Octubre 2018.

- Segura-Martínez, M. T., L. O. Tejada, E. Ruiz-Cancino, y J. H. Silva-Espinosa. 1995. Evaluación de dos insecticidas para el control de adultos de la mosca mexicana de la fruta. La revista de la UAT. 41: 57-59. https://www.researchgate.net/publication/281638156_Evaluacion_de_dos_insecticidas_para_el_control_de_adultos_de_la_mosca_mexicana_de_la_fruta
- Vilatuña, J., P. Valenzuela, J. Bolaños, R. Hidalgo, and A. Mariño. 2016. Hosts of fruit flies *Anastrepha* spp. and *Ceratitis Capitata* (Diptera: Tephritidae) in Ecuador. Revista Científica Ecuatoriana. <https://doi.org/10.36331/revista.v3i1.16>

Natural Enemies of Sugarcane Aphid, *Melanaphis sacchari* Zehntner¹, in Southern Tamaulipas, Mexico

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Abstract. Sugarcane aphid, *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae), can greatly reduce yield potential of sorghum, *Sorghum bicolor* (L.) Moench. Sugarcane aphid and its natural enemies were sampled in pesticide-free fields at Altamira, Tamaulipas, Mexico, during a 2-year period. Seven species of predators identified were: *Orius* sp. Wolff (Hemiptera: Anthocoridae), *Cycloneda sanguinea* (L.), *Hippodamia convergens* Guérin-Méneville, and *Scymnus* sp. Kugelann (Coleoptera: Coccinellidae); *Chrysoperla* sp. Steinmann (Neuroptera: Chrysopidae); and *Eosalpingogaster* sp. Hull, and *Eupeodes* sp. Osten-Sacken (Diptera: Syrphidae). Five parasitoid species associated with the pest were *Pachyneuron aphidis* Bouché and *Pachyneuron muscarum* L. (Hymenoptera: Pteromalidae), *Lysiphlebus testaceipes* Cresson (Hymenoptera: Braconidae), and *Aphelinus mali* Haldeman and *Aphelinus varipes* Förster (Hymenoptera: Aphelinidae). *Pachyneuron* spp. parasitized 68.25%, while other parasitoids parasitized 6.44% of the aphids sampled. This research furthers understanding of already existing and ongoing parasitism/predation and enriches knowledge of natural enemies that can be used as biological control agents against *M. sacchari*. *A. mali*, *P. aphidis*, and *P. muscarum* were reported as parasitoids of *M. sacchari*.

Introduction

Forty percent of sorghum, *Sorghum bicolor* (L.) Moench (Poaceae), in Mexico is produced in the State of Tamaulipas (SIAP 2022). Sugarcane aphid, *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae), is a common pest of sorghum in tropical Africa, Asia, and the southern United States. Sugarcane aphid infests the underside of leaves, removes plant sap from xylem of leaves, induces red spots or streaks, and when abundant causes physiological loss such as wilting/curling of leaves and chlorosis and deposits of sugary substances on the surface of leaves, which leads to

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development of sooty mold fungi. Quality of grain and yield at harvest are less (Singh et al. 2004, SENASICA 2014).

In late 2013, *M. sacchari* was detected causing major loss at Tamaulipas (Maya-Hernández and Rodríguez-del-Bosque 2014). Numbers of aphids necessary to cause yield reduction in sorghum vary based on plant stage, interval between and duration of infestation (Singh et al. 2004). Yield loss varies from 30 to 100% if effective control is not used, with 10% average yield loss for each leaf damaged (Rodríguez-del-Bosque and Teran 2015).

Currently, the aphid is in the Mexican states of Coahuila, Guanajuato, Jalisco, Michoacán, Nayarit, Nuevo León, San Luis Potosí, Sinaloa, and Tamaulipas (Peña-Martínez et al. 2018). Strategies to control sugarcane aphid include elimination of alternate hosts, conservation of natural enemies, planting resistant hybrids of sorghum (Quijano-Carranza et al. 2017), and application of insecticide (CESAVEG 2017).

Singh et al. (2004) listed more than 47 species of Coleoptera, Diptera, Hemiptera, Hymenoptera, and Neuroptera that are natural enemies of *M. sacchari*. Predators (Cortez-Mondaca et al. 2016, Provisor-Bermúdez and López-Martínez 2016, Rodríguez-Palomera et al. 2016, Rodríguez-Vélez et al. 2016) and parasitoids (López-Gutiérrez et al. 2016, García-González et al. 2018) associated with *M. sacchari* occur in different regions of Mexico. Although there are several studies of parasitoid and predator species associated with sugarcane aphid in Mexico, only three studies of natural enemies were associated with the pest in Tamaulipas State (Rodríguez-Vélez et al. 2016, 2019; Rodríguez-del-Bosque et al. 2018), and there is only one study in southern Tamaulipas (Rodríguez-Vélez et al. 2019). Identification of natural enemies is important for development of effective integrated pest management. The objective of this research was to identify natural enemies of sugarcane aphid at Altamira, Tamaulipas, Mexico.

Materials and Methods

Sorghum crops (var. DOW 1445) were sampled in five fields at Las Huastecas Research Station of Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP) ($22^{\circ}34'26.6''N$, $98^{\circ}10'12.4''W$; $22^{\circ}34'08.4''N$, $98^{\circ}10'04.2''W$; $22^{\circ}34'06.3''N$, $98^{\circ}10'29.7''W$; $22^{\circ}33'56.3''N$, $98^{\circ}10'15.8''W$, and $22^{\circ}33'17.7''N$, $98^{\circ}09'40.8''W$) at Estación Cuauhtémoc, Altamira, Tamaulipas, Mexico. Sampling was during April (before harvest in June and the next crop is sown in July) and from August to November in 2018 and from February to August 2019, with sampling every 35 days.

Direct sampling and visual inspections were made of predator adult insects. Sampling was with a paintbrush and entomological tweezers at five points chosen at random in each field. Ten plants per sampling point were randomly chosen and carefully examining from the base to the youngest leaf or spike. Insects collected were preserved in 75% ethanol and labeled with location and date. Species identified in the field were counted and recorded, and species identified were confirmed in a laboratory.

Syrphid larvae were collected live and reared to adulthood on *M. sacchari* for identification in a laboratory. Parasitoids were sampled at each date at five sampling points by collecting an entire leaf of sorghum from the middle of a plant infested with *M. sacchari* with and without parasitism symptoms (mummification and color change). Infested leaves were transferred in brown paper bags to an environmental chamber

(Lab-Line Biotronette Mark III model 846) in the Departamento de Parasitología (Plant Health Department) at the Universidad Autónoma Agraria Antonio Narro (UAAAAN), Saltillo, Coahuila, Mexico. Leaves with mummified aphids were treated using the method in Bodlah et al. (2012). The method kept aphids alive to allow development of parasitoids. A leaf was broken into small sections and put with parasitized aphids into containers covered with museline to allow gas exchange. Emergence of parasitoids was monitored every day for 2 months after each sample was taken. Emerged parasitoids were collected and transferred into 75% ethanol in 2-ml vials for counting, preservation, and taxonomic identification.

Adult aphids that underwent discoloration with 40% potassium hydroxide (KOH) solution were permanently preserved using Canada balsam mounted on slides (Peña-Martínez et al. 2000) and identified using keys by Blackman and Eastop (2006) and Rodríguez-del-Bosque and Terán-Vargas (2018). Predators were identified to family using Triplehorn and Johnson (2005) and Herring (1976) and to species using Thompson (1999), Valencia-Luna et al. (2006), and Larson (2013) with the aid of a stereomicroscope. Parasitoids were permanently preserved by methodology described in Noyes (1982) and identified to family using Triplehorn and Johnson (2005) and to species using Graham (1969), Gibson et al. (1997), Wharton et al. (1997), Fernández and Sharkey (2006), Narendran et al. (2007), Myartseva et al. (2012), Zamora-Mejías and Hanson (2017), and Tomanović et al. (2018). Parasitized aphids were dissected to inspect the primary or secondary parasitoid. Specimens were curated in the Departamento de Parasitología of the UAAAAN. Percentage of parasitism of each species was calculated by using the formula of Hunsberger and Peña (1997) as a fraction of the total parasitoids collected from the total population of aphids sampled:

$$\text{Parasitism (\%)} = \frac{\text{Number of parasitoids emerged}}{\text{Number of parasitoids emerged} + \text{Number of adult hosts}} * 100$$

Results and Discussion

Aphids in the sampled fields were confirmed as *M. sacchari* characterized by yellow color with gray or brown tones, dark tarsi, and siphunculi (Rodríguez-del-Bosque and Terán-Vargas 2018). Seven species of predators in five families were identified: *Orius* sp. Wolff (Hemiptera: Anthocoridae), *Cycloneda sanguinea* (L.), *Hippodamia convergens* Guérin-Méneville and *Scymnus* sp. Kugelann (Coleoptera: Coccinellidae), *Eosalpingogaster* sp. Hull and *Eupeodes* sp. Osten-Sacken (Diptera: Syrphidae), and *Chrysoperla* sp. Steinmann (Neuroptera: Chrysopidae). *Orius* sp. with four specimens collected was the only Hemiptera associated with *M. sacchari* in the sampled area. Bowling et al. (2016) and Maxson et al. (2019) also reported *Orius insidiosus* Say associated with sugarcane aphid. Although not found in this study, Singh et al. (2004) documented the Hemiptera species anthocorid *Scoloposcelis parallelus* Mostch and *Geocoris* sp. Fallén (Geocoridae) as sugarcane aphid predators.

Coccinellids *C. sanguinea*, *H. convergens*, and *Scymnus* spp. were reported as predators of *M. sacchari* by Singh et al. (2004), Bowling et al. (2016), Maxson et al. (2019), and Farias et al. (2022). In Mexico, Provisor-Bermúdez and López-Martínez (2016) reported the first two species at Morelos, and Rodríguez-Palomera et al. (2016) and Rodríguez-Vélez et al. (2016) found all three species associated with *M. sacchari* at Nayarit and Tamaulipas. Cortez-Mondaca et al. (2016) reported

three in northern Sinaloa. Rodríguez-del-Bosque et al. (2018) reported 11 species of coccinellids feeding on *M. sacchari*. We collected 25 adult specimens of *C. sanguinea*, 32 adults of *H. convergens*, and 73 adults of *Scymnus* sp.; this last was the most frequent and abundant species.

From February to March 2019, five syrphid larvae were collected and reared to adulthood. Two syrphid species were identified as *Eosalpingogaster* sp. and the remaining specimens as *Eupeodes* sp. According to Mengual and Thompson (2011), larvae of *Eosalpingogaster* are predators of Sternorrhyncha hemipterans. Some *Eupeodes* species are important in ecosystems where adults are pollinators and larvae are biological control agents of aphids in various crops (Turk et al. 2015). Cortez-Mondaca et al. (2016) reported *Allograpta obliqua* (Say) and *Eupeodes americanus* (Wiedemann) associated with sugarcane aphids at Sinaloa. Rodríguez-Del-Bosque et al. (2018) reported the syrphid *A. obliqua* feeding on *M. sacchari* in northern Tamaulipas.

Only three adult specimens of green lacewings of the genus *Chrysoperla*, potential generalist predator of many arthropods such as sugarcane aphids, were collected in April 2018 and one in February 2019. In northern Sinaloa, México, Cortez-Mondaca et al. (2016) found eight species associated with *M. sacchari*: *Ceraeochrysa caligata* Banks, *Ceraeochrysa cubana* Hagen, *Ceraeochrysa* sp. nr *cincta* Schneider, *Ceraeochrysa valida* Banks, *Chrysoperla carnea* Stephens, *Chrysoperla comanche* Banks, *Chrysoperla rufilabris* Burmeister, and *Chrysoperla externa* Hagen; this last was most abundant. Chrysopids *C. valida* and *Chrysoperla* sp. (*carnea* group) were recorded at Tamaulipas by Rodríguez-del-Bosque et al. (2018). Maxson et al. (2019) observed five green lacewing species.

Of 5,421 aphids collected during 10 sample dates, 74.69% were parasitized by five species of Hymenoptera: *Lysiphlebus testaceipes* Cresson (Braconidae), *Pachyneuron aphidis* Bouché and *Pachyneuron muscarum* L. (Pteromalidae), and *Aphelinus mali* Haldeman and *Aphelinus varipes* Förster (Aphelinidae). *L. testaceipes* was found in April and October 2018 and from February to May 2019, parasitizing 6.07% of *M. sacchari* collected. *L. testaceipes* was reported as a parasitoid of *M. sacchari* by Singh et al. (2004), Bowling et al. (2016), Maxson et al. (2019), and Giles et al. (2021), and in Mexico by López-Gutiérrez et al. (2016), García-González et al. (2018), Rodríguez-del-Bosque et al. (2018), and Rodríguez-Vélez et al. (2019). López-Gutiérrez et al. (2016) found the parasitoid associated with four other genera of Braconidae: Aphidiinae (*Aphidius* Esenbeck, *Praon* Haliday, *Binodoxys* Mackauer, and *Diaretiella* Stary) in populations of *M. sacchari*, but point out they were not enough to reduce aphid abundance below the economic threshold at Irapuato, Guanajuato, Mexico. A probable reason for less control was that *L. testaceipes* larvae are affected by *Hamiltonella defensa* Moran et al. (Enterobacterales: Enterobacteriaceae), a bacterial symbiont of several aphid species that protects the aphid host against parasitism by braconid wasps of the subfamily Aphidiinae. Yet, no relevant test determined if aphids at Altamira, Tamaulipas, possessed the symbiosis or if local *L. testaceipes* were resistant to the bacteria. Lahiri et al. (2019) suggested that *L. testaceipes* was not well synchronized with occurrence of *M. sacchari*, and additional natural enemies (e.g., other parasitoids and/or predators) are necessary to manage the pest.

Pachyneuron aphidis and *P. muscarum* were collected at each sampling time, except in July 2019, because of absence of aphids. Both species parasitized 68.25% of the aphids and were the only species associated with *M. sacchari* when the aphid was not abundant.

In total, 2,269 *P. aphidis* and 1,431 *P. muscarum* were collected. *Pachyneuron* spp. Walker has cosmopolitan distribution, with most species being primary parasitoids and some hyperparasitoids of Sternorrhyncha Hemiptera (Kfir and Rosen 1981). Aphids with hyperparasitoids were Aphididae, *Aphis gossypii* (Glover), *Lipaphis erysimi* (Kaltenbach), *Aphis craccivora* Koch, *Rhopalosiphum maidis* (Fitch), and *Myzus persicae* (Sulzer) (Ceballos et al. 2009). Hyperparasitism of these genera were by *Pachyneuron crassiculme* Waterston that emerged from parasitized *Diaphorina citri* Kuwayama (Hemiptera: Liviidae) nymphs by *Diaphorencyrtus aligarhensis* (Shafee, Alam, and Agarwal) (Hymenoptera: Encyrtidae), and *Tamarixia radiata* (Waterston) (Hymenoptera: Eulophidae) (Bistline-East and Hoddle 2014).

P. muscarum is a hyperparasitoid of various primary parasitoids on *Aphis* sp. L., *Sitobion avenae* F., *R. maidis*, *Rhopalosiphum padi* L., and *P. aphidis* of *Brevicoryne brassicae* (L.), *L. erysimi*, *Aphis fabae* Scopoli, *Acyrthosiphon pisum* (Harris), *S. avenae*, *R. maidis*, and *R. padi* (Kos et al. 2012). *P. aphidis* was reported by García-González et al. (2018) and Payán-Arzápalo et al. (2018) as a hyperparasitoid of *M. sacchari*, but they did not indicate which primary parasitoid was the host. Rodríguez-del-Bosque et al. (2018) and Rodríguez-Vélez et al. (2019) reported *P. aphidis* as a hyperparasitoid of *L. testaceipes* associated with *M. sacchari* at Tamaulipas, Mexico. However, the Universal Chalcidoidea Database (Noyes 2022) indicated *P. muscarum* and *P. aphidis* were primary parasitoids of a diversity of aphid species including *M. sacchari* as a primary host of *P. aphidis*.

We believe *P. aphidis* and *P. muscarum* are not hyperparasitoids of sugarcane aphid for two reasons. Abundance of *Pachyneuron* was greater than that of braconids at all sampling dates except October 2018, indicating *Pachyneuron* spp. parasitized *M. sacchari* when other species were absent. We also found hyperparasitoid *Alloxysta* sp. Förster (Hymenoptera: Encyrtidae), and dissection indicated the number of hyperparasitized aphids coincided with the amount of *Alloxysta* sp. collected. DeBach (1964) indicated that when dissecting a hyperparasitized host, the meconium and exuvium of the primary parasitoid would be found. We observed this during dissection of aphids hyperparasitized and parasitized by *Alloxysta* sp. and *L. testaceipes*. When dissecting mummies where *P. aphidis* and *P. muscarum* had emerged, however, only meconia were found, corroborating the roles of the two species as primary parasitoids.

Fifteen specimens of *A. varipes* with 0.28% parasitism rate were collected in August 2019. Five specimens of *A. mali* were collected in May 2019, with 0.09% parasitism. Thus far, *A. mali* was not reported associated with sugarcane aphids.

Singh et al. (2004) reported *A. maidis* associated with *M. sacchari*. However, in Texas, Maxson et al. (2019) reported *A. varipes* and *Aphelinus nigritus* Howard (Aphelinidae) parasitizing the aphid, in northern Tamaulipas Rodríguez-del-Bosque et al. (2018) reported an *Aphelinus* sp., and at Coahuila, Mexico, García-González et al. (2018) reported *A. varipes* and *L. testaceipes* associated with *M. sacchari*.

Giles et al. (2021) documented that *A. nigritus* was relatively abundant parasitizing sugarcane aphids in winter crops during late fall and early spring in Oklahoma. Farias et al. (2022) reported the parasitoid represented 90% of the natural enemies of *M. sacchari* on the Texas Gulf Coast and in Central Oklahoma.

This research furthers understanding of already existing and ongoing parasitism/predation, provides information about beneficial entomofauna associated with sugarcane aphid in southern Tamaulipas, and increases knowledge of natural enemies that have potential for biological control of this pest or even in an integrated management program for *M. sacchari*. The study is the first report of *A. mali* as a

parasitoid of *M. sacchari* at Altamira, Tamaulipas. *P. aphidis* and *P. muscarum* were reported as parasitoids of *M. sacchari*. Additional work is needed to confirm *Pachyneuron* spp. as primary parasitoids of *M. sacchari*, quantify effects on *M. sacchari* populations, and identify other hosts.

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References Cited

- Bistline-East, A., and M. S. Hoddle. 2014. *Chartocerus* sp. (Hymenoptera: Signiphoridae) and *Pachyneuron crassiculme* (Hymenoptera: Pteromalidae) are obligate hyperparasitoids of *Diaphorencyrtus aligarhensis* (Hymenoptera: Encyrtidae) and possibly *Tamarixia radiata* (Hymenoptera: Eulophidae). *Fla. Entomol.* 97: 562-566.
- Blackman, R. L., and V. F. Eastop. 2006. Aphids on the World's Herbaceous Plants and Shrubs. John Wiley & Sons.
- Bodlah, I., M. Naeem, and A. Mohsin. 2012. Distribution, hosts and biology of *Diaeletiella rapae* (McIntosh) (Hymenoptera: Braconidae: Aphidiinae) in Punjab, Pakistan. *Pak. J. Zool.* 44: 1307-1315.
- Bowling, R. D., M. J. Brewer, D. L. Kerns, J. Gordy, N. Seiter, N. E. Elliott, G. D. Buntin, M. O. Way, T. A. Royer, S. Biles, and E. Maxson. 2016. Sugarcane aphid (Hemiptera: Aphididae): a new pest on sorghum in North America. *J. Integr. Pest Manag.* 7: 1-13.
- Ceballos, M., M. A. Martínez, L. Duarte, H. L. Baños, y A. Sánchez. 2009. Asociación áfidos-parasitoides en cultivos hortícolas. *Rev. Prot. Veg.* 24: 180-183.
- CESAVEG (Comité Estatal de Sanidad Vegetal de Guanajuato). 2017. Guía para el manejo del pulgón amarillo del sorgo. México. buenas_practicas_agricolas/Guia_para_el_manejo_del_pulgón_amarillo.pdf Accessed 7 April 2022.
- Cortez-Mondaca, E., M. López-Buitimea, J. I. López-Arroyo, F. J. Orduño-Cota, and G. Herrera-Rodríguez. 2016. Especies de Chrysopidae asociadas al pulgón del sorgo en el norte de Sinaloa, México. *Southwest. Entomol.* 41: 541-545.
- DeBach, P. 1964. Biological Control of Insect Pests and Weeds. Chapman & Hall.
- Farias, A. M., N. C. Elliott, and M. J. Brewer. 2022. Suppression of the sugarcane aphid, *Melanaphis sacchari* (Hemiptera: Aphididae), by resident natural enemies on susceptible and resistant sorghum hybrids. *Environ Entomol.* nvab147. doi: 10.1093/ee/nvab147.
- Fernández, F., and M. J. Sharkey. 2006. Introducción a los Hymenoptera de la región Neotropical. Universidad Nacional de Colombia. Bogotá, Colombia.
- García-González, F., M. Ramírez-Delgado, H. Cortez-Madrigal, and M. L. Ramírez-Ahuja. 2018. New reports of parasitoids and hyperparasitoids of *Melanaphis sacchari* (Zehntner) in Mexico. *Southwest. Entomol.* 43: 787-790.
- Gibson, G. A. P., I. T. Huber, and J. B. Woolley. 1997. Annotated Keys to the Genera of Nearctic Chalcidoidea (Hymenoptera). NRC Research Press.

- Giles, K. L., N. C. Elliott, H. E. Butler, K. A. Baum, and G. F. Backoulou. 2021. Increase in importance of *Aphelinus nigritus* (Howard) on winter crops in Oklahoma coincides with invasion of sugarcane aphid on sorghum in Oklahoma. Southwest. Entomol. 46: 59-68.
- Graham, M. W. R. De Vere. 1969. The Pteromalidae of North-Western Europe (Hymenoptera: Chalcidoidea). Bull. Brit. Mus. (Natural History) Entomology.
- Herring, J. L. 1976. Keys to genera of Anthocoridae of America north of Mexico, with description of a new genus (Hemiptera: Heteroptera). Fla. Entomol. 59: 143-150.
- Hunsberger, G. B. A., and E. J. Peña. 1997. *Catolaccus hunteri* (Hymenoptera: Pteromalidae), a parasite of *Anthonomus macromalus* (Coleoptera: Curculionidae) in South Florida. Fla. Entomol. 80: 301-304.
- Kfir, R., and D. Rosen. 1981. Biology of the hyperparasite *Pachyneuron concolor* (Förster) (Hymenoptera: Pteromalidae) reared on *Microterys flavus* (Howard) in brown soft scale. J. Entomol. Soc. South Afr. 44: 151-163.
- Kos, K., O. Petrović-Obradović, V. Žikić, A. Petrović, S. Trdan, and Ž. Tomanović. 2012. Review of interactions between host plants, aphids, primary parasitoids and hyperparasitoids in vegetable and cereal ecosystems in Slovenia. J. Entomol. Res. Soc. 14: 67-78.
- Lahiri, S., N. Xinzhi, G. D. Buntin, and M. D. Toews. 2019. Parasitism of *Melanaphis sacchari* (Hemiptera: Aphididae) by *Lysiphlebus testaceipes* (Hymenoptera: Braconidae) in the greenhouse and field. J. Entomol. Sci. 55: 14-24.
- Larson, D. J. 2013. Key to lady beetles (Coleoptera: Coccinellidae) of Saskatchewan. Entomol. Soc. Saskatchewan. http://www.entsocsask.ca/documents/insect_lists/Coccinellidae_key.pdf Accessed 10 March 2020.
- López-Gutiérrez, D. R., M. D. Salas-Araiza, O. A. Martínez-Jaime, and E. Salazar-Solís. 2016. Géneros de Aphidiidae (Hymenoptera) parasitando al pulgón amarillo de la caña de azúcar *Melanaphis sacchari* Zehntner, 1897 (Hemiptera: Aphididae) en Irapuato, Guanajuato, México. Entomol. Mex. 3: 365-368. <http://www.acaentmex.org/entomologia/revista/2016/EA/Em%20365-368.pdf> Accessed 20 January 2022.
- Maxson, E. L., M. J. Brewer, W. L. Rooney, and J. B. Wooley. 2019. Species composition and abundance of the natural enemies of sugarcane aphid, *Melanaphis sacchari* (Zehntner) (Hemiptera: Aphididae), on sorghum in Texas. Proc. Entomol. Soc. Wash. 121: 657-680.
- Maya-Hernández, V., and L. A. Rodríguez-del-Bosque. 2014. Pulgón amarillo: una nueva plaga del sorgo en Tamaulipas. Centro de Investigación Regional Noreste, Campo Experimental Río Bravo. Desplegable para productores. INIFAP/CIRNE: A-532. <http://www.inifapcirne.gob.mx/Biblioteca/Publicaciones/983.pdf> Accessed 15 April 2022.
- Mengual, X., and F. C. Thompson. 2011. Carmine cochineal killers: the flower fly genus *Eosalpingogaster* Hull (Diptera: Syrphidae) revised. Syst. Entomol. 36: 713-731.
- Myartseva, S. N., E. Ruíz-Cancino, and J. M. Coronado-Blanco. 2012. Aphelinidae (Hymenoptera: Chalcidoidea) de importancia agrícola en México. Revisión y claves. Departamento de Fomento Editorial de la UAT. Ciudad Victoria, Tamaulipas, México. <https://riuat.uat.edu.mx/handle/123456789/1561> Accessed 20 February 2020.

- Narendran, T. C., S. Santhosh, P. Abhilash, M. Sheeba, and M. C. Jiley. 2007. A review of *Pachyneuron* species (Hymenoptera: Pteromalidae) of Middle East. *J. Environ. Sociobiol.* 4: 119-138.
- Noyes, J. S. 1982. Collecting and preserving chalcid wasps (Hymenoptera: Chalcidoidea). *J. Nat. Hist.* 16: 315-334.
- Noyes, J. S. 2022. Universal Chalcidoidea Database. World Wide Web electronic publication. <http://www.nhm.ac.uk/chalcidoids> Accessed 17 April 2022.
- Payán-Arzapalo, M. A., C. E. Ail-Catzim, R. Gastélum-Luque, J. E. Guerra-Liera, M. G. Yáñez-Juárez, and M. L. Ramírez-Ahuja. 2018. Parasitism and hyperparasitism in *Melanaphis sacchari* Zehntner in sorghum in Mexico. Southwest. Entomol. 43: 433-437.
- Peña-Martínez, R., N. Villegas-Jiménez, and J. R. Lomelí-Flores. 2000. Técnicas de colecta, montaje y preservación de pulgones y algunos de sus enemigos naturales. In H. C. Arredondo-Bernal, M. A. Mellín-Rosas, P. Pérez-Serrato, and J. P. Martínez-Soriano [eds.], Control Biológico del Pulgón Café *Toxoptera citricida*, Vector del Virus de la Tristeza de los Cítricos.
- Peña-Martínez, R., J. R Lomelí-Flores, R. Bujanos-Muñiz, A. L. Muñoz-Viveros, J. M. Venegas-Rico, R. Salas Monzón, O. E. Hernández-Torres, A. Marín-Jarillo, and J. E. Ibarra-Rendón. 2018. Pulgón amarillo del sorgo (PAS), *Melanaphis sacchari* (Zehntner, 1897), interrogantes biológicas y tablas de vida. Fundación Guanajuato Produce A.C.
- Provisor-Bermúdez, Y., and V. López Martínez. 2016. Primer registro de *Hippodamia convergens* y *Cycloneda sanguínea* (Coleoptera: Coccinellidae), como depredadores de *Melanaphis sacchari* (Hemiptera: Aphididae), en sorgo, en Morelos, México. Acta Agríc. Pecuaria 2: 51-53.
- Quijano-Carranza, J. A., V. Q. Pecina, M. R. Bujanos, J. A. Marín, and L. R. Yáñez. 2017. Guía para el manejo del pulgón amarillo del sorgo. Folleto para productores No. 1. Fundación Guanajuato Produce A. C. https://issuu.com/sdayr/docs/cuadernillo_pulgo_n_alta_para_impr Accessed 20 September 2021.
- Rodríguez-del-Bosque, L. A., and A. P. Terán. 2015. *Melanaphis sacchari* (Hemiptera: Aphididae): a new sorghum insect pest in Mexico. Southwest. Entomol. 40: 433-434.
- Rodríguez-del-Bosque, L. A., and A. P. Terán-Vargas. 2018. Manejo integrado del pulgón amarillo del sorgo en Tamaulipas. Centro de Investigación Regional del Noreste, Campo Experimental Río Bravo, Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias. https://www.gob.mx/cms/uploads/attachment/file/394239/Manejo_integrado_del_pulg_n_amarillo_del_sorgo_en_Tamaulipas.pdf Accessed 15 April 2022.
- Rodríguez-del-Bosque, L. A., B. Rodríguez-Vélez, M. A. Sarmiento-Cordero, and H. C. Arredondo-Bernal. 2018. Natural enemies of *Melanaphis sacchari* on grain sorghum in northeastern Mexico. Southwest. Entomol. 43: 277-279.
- Rodríguez-Palomera, M., J. Cambero-Campos, G. Luna-Esquível, O. Estrada-Virgen, N. De Dios-Ávila, and C. Cambero-Ayón. 2016. Coccinélidos depredadores del pulgón amarillo del sorgo *Melanaphis sacchari* (Zehntner) (Hemiptera: Aphididae) en Nayarit, México. Entomol. Mex. 3: 361-364.
- Rodríguez-Vélez, B., A. Suaste-Dzul, A. Gallou, J. M. Rodríguez-Vélez, M. A. Sarmiento-Cordero, and H. C. Arredondo-Bernal. 2019. Pulgones (Hemiptera: Aphididae) y sus parasitoídes (Hymenoptera) en cultivos de sorgo en los estados de Colima y Tamaulipas, México. Acta Zool. Mex. (n.s.) 35: 1-9.

- Rodríguez-Vélez, J. M., B. Rodríguez-Vélez, M. A. Sarmiento-Cordero, M. Palomares-Pérez, and H. C. Arredondo-Bernal. 2016. Species of Coccinellidae (Coleoptera: Cucujoidea) associated with *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae) in Tamaulipas, Mexico. Entomol. News 126: 97-105.
- SENASICA (Servicio Nacional de Sanidad, Inocuidad y Calidad Agroalimentaria). 2014. Pulgón amarillo *Melanaphis sacchari* (Zehntner). Ficha Técnica 43. http://www.cesavep.org/descargas/SORGO/FichaT%C3%A9cnicaNo.43Pulg onamarillo_25junio.pdf. Accessed 15 April 2022
- SIAP (Servicio de Información Agroalimentaria y Pesquera). 2022. Cierre de la Producción Agrícola. Anuario Estadístico de la Producción Agrícola. México. <https://nube.siap.gob.mx/cierreagricola> Accessed 16 April 2022.
- Singh, B. U., P. G. Padmaja, and N. Seetharama. 2004. Biology and management of the sugarcane aphid, *Melanaphis sacchari* (Zehntner) (Homoptera: Aphididae), in sorghum: a review. Crop Prot. 23: 739-755.
- Thompson, F. C. 1999. A key to the genera of the flower flies (Diptera: Syrphidae) of the Neotropical Region including descriptions of new genera and species and a glossary of taxonomic terms, pp. 321-378. In V. K. Gupta [ed.], Contributions on Entomology, International. Vol. 3. Associated Publishers.
- Tomanović, Ž., M. Mitrović, A. Petrović, N. G. Kavallieratos, V. Žikić, A. Ivanović, E. Rakhshani, P. Starý, and C. Vorburger. 2018. Revision of the European *Lysiphlebus* species (Hymenoptera: Braconidae: Aphidiinae) on the basis of COI and 28SD2 molecular markers and morphology. Arthropod Syst. Phylogeny 76: 179-213.
- Triplehorn, C. A., and N. F. Johnson. 2005. Introduction to the Study of Insects. 7th edition. Thompson Brooks/Cole.
- Turk, J. K., N. Memon, B. Mal, S. A. Memon, M. A. Shah, and D. A. Solangi. 2015. Redescription of two species of genus *Eupeodes* Osten Sacken from Quetta Balochistan, Pakistán. J. Anim. Plant Sci. 25: 1329-1334.
- Valencia-Luna, L. A., J. Romero-Nápoles, J. Valdez-Carrasco, J. L. Carrillo-Sánchez, y V. López-Martínez. 2006. Taxonomía y registros de Chrysopidae (Insecta: Neuroptera) en el estado de Morelos, México. Acta Zool. Mex. (n.s.) 22: 17-61.
- Wharton, R. A., P. M. Marsh, and M. J. Sharkey. 1997. Manual of the new world genera of the family Braconidae (Hymenoptera). Int. Soc. Hymenopterists.
- Zamora-Mejías, D., and P. E. Hanson. 2017. Clave dicotómica para especies parásitoides e hiperparásitoides (Hymenoptera) de áfidos (Hemiptera: Aphididae) de Costa Rica. Agron. Mesoam. 28: 565-572.

New Record of *Anisopteromalus calandrae*¹ at Veracruz, Mexico: A Parasitoid of *Sitophilus zeamais*²

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Abstract. *Anisopteromalus calandrae* from Veracruz, Mexico, was characterized morphologically and molecularly. Specimens associated with *Sitophilus zeamais* (Motschulsky) in stored maize (*Zea mays* L) were collected. Morphology and the barcode region of the cytochrome oxidase 1 confirmed the identity of the parasitoid.

Introduction

Anisopteromalus calandrae (Hymenoptera: Pteromalidae) is a solitary ectoparasitic wasp of the maize weevil, *Sitophilus zeamais* (Motschulsky) (Coleoptera: Curculionidae), and rice weevil, *S. oryzae* (L.) (Schöller et al. 2006, Ghimire and Phillips 2008). This species of wasp is one of the most studied for its potential as a parasitoid of stored grain weevils (Belda and Riudavets 2011, Adarkwah et al. 2014). Several studies of *A. calandrae* described its use as a biological control agent, its life cycle, and susceptibility and resistance to insecticides (Baker et al. 1998). However, knowledge about its taxonomy and systematics is limited, so it is necessary to find other techniques that complement identification (Baur et al. 2014).

Seven species of *Anisopteromalus* were reported worldwide [*A. apiovorus* Rasplus, *A. calandrae* (Howard), *A. caryedophagus* Rasplus, *A. ceylonensis* Sureshan, *A. cornis* Baur, *A. indicus* Gupta, and Sureshan and *A. quinarius* Gokhman and Baur] (Baur et al. 2014, Gupta and Sureshan 2014). The species are distributed mainly in the Old World (Fig. 1) and are parasitoids of larvae of Lepidoptera and Coleoptera (Noyes 2022).

Maize is a crop of economic importance because of its use for human consumption and provision of raw materials for the food industry. In 2020, about 157.2 million tons were produced worldwide (FAO 2020). But, 20% made is lost because of the activity of insect pests in stored grain (Pingali and Pandey 2001). In

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for 86% of production, mainly destined for human consumption (SIAP 2017). Depending on the area and climatic region, maize storage losses range from 10 to 40%, and much of the loss is attributed to insect pests, of which *S. zeamais* is the most important (Tigar et al. 1994, García-Lara and Bergvinson 2007). Several physical, chemical, and biological methods are used to control the pest. The most common methods are based on the use of synthetic insecticides; however, disadvantages include pest resistance, environmental contamination, and human health risks. For these reasons, it is important to include biological control as a priority to reduce the impact of *S. zeamais* on stored grain (Mahal et al. 2005).

A. calandrae in Mexico has been reported in the literature review as a parasitoid of *S. zeamais* (Noyes 2022). Still, its presence has not been confirmed, and there are no distribution data for the country. We report *A. calandrae* at Veracruz, Mexico. We identified *A. calandrae* by morphology and sequencing a portion of the Cytochrome Oxidase 1 gene (CO1). The results will facilitate and complement taxonomic studies for the identification of *A. calandrae*.



Fig. 1. Distribution report of the *Anisopteromalus* species worldwide (Noyes 2022).

Materials and Methods

Specimens were collected in May 2017 at José Azueta city ($18^{\circ}04'02.2''$ N $95^{\circ}42'44.0''$ W) in the State of Veracruz, Mexico, from stored kernels of maize infested with *S. zeamais*. Adult specimens were transported to the laboratory of Fisiología Molecular y Estructural (FCB-UANL) and put into 96% ethanol for morphological and molecular analyses. Voucher specimens were deposited in the “Colección de

Insectos Benéficos Entomófagos" (CIBE 17-001) (FCB-UANL). The Qiagen DNeasy kit (Hilden, Germany) was used as described by Giantsis et al. (2016) to nondestructively isolate genomic DNA from the whole specimen (5♀ 5♂). Polymerase chain reaction (PCR) was used to amplify the DNA barcode region of the Cytochrome Oxidase Subunit 1 (CO1) using LCO1490fw (5'-GGTCAACAAATCATAAAGATATTGG-3') and HCO2198rev (5'-TAAACTTCAGGGTGACCAAAAAATCA-3') primers (Folmer et al. 1994). PCRs were done in 20- μ l reaction volume: 2 μ l of DNA, 2 μ l of 10X Qiagen PCR buffer containing 15 mM MgCl₂, 0.9 μ l of each primer, 0.6 μ l of dNTPs (25 mM each), and 0.2 μ l of (5 U/ μ l) Taq DNA Polymerase (Qiagen, Hilden, Germany), and 13.4 μ l of H₂O. PCR conditions were: 94°C for 3 minutes, followed by 40 cycles of 94°C for 30 seconds, 52°C for 1 minute, 72°C for 1 minute with final extension at 72°C for 10 minutes. All PCR products were electrophoresed through an agarose gel (1%). DNA was sequenced on an ABI 3100 Genetic Analyzer, using Big Dye termination version 3.1, both from Life Technologies. GeneStudio Pro software suite (GeneStudio, Suwanee, GO) computer programs were used to edit for quality sequencing chromatograms that were trimmed to remove sequences of primers. Amplicons resulting from PCRs of 10 organisms were sequenced. DNA sequences of *A. calandrae* for comparison were downloaded from BOLD and NCBI databases. The sequences obtained in the study were deposited as accession number MK272965 in GenBank.

Results

In total, 142 specimens were examined and determined as *A. calandrae* based on keys by Baur et al. (2014). Seventy-five females (Fig. 2A) and 67 males (Fig. 2B) were analyzed. The first and second funicular segment of females was the same size (Fig. 2C), while the first funicular segment of males always was shorter than the second funicular segment (Fig. 2D). A product of 651 bp corresponding to the CO1 gene was amplified, and bioinformatic analyses of the obtained sequences confirmed the identity of the species as *A. calandrae* (GenBank accession MK272965). CO1 sequences were identical among females and males sampled from Veracruz, and more than 99% of the pairs of bases were identical to a series of sequences labeled as *A. calandrae* in the GenBank database. By comparison with previously reported sequences of the same species, seven changes were found, from which only one corresponded to a non-synonymous change (Table 1).

Discussion

Insects with an estimated 10⁷ species are the most abundant of all life on earth (Pons et al. 2006). For identification, morphology-based taxonomy has limitations such as unclear species recognition and unrecognized cryptic species. In this study, morphological and molecular analyses confirmed *Anisopteromalus calandrae* parasitizing *S. zeamais* larvae at Veracruz, Mexico. In Mexico, the parasitoid species was once reported to parasitize *S. zeamais* (Noyes 2022), but until now, its identity was not confirmed.

Use of genomic identification systems to identify species has been used for a wide variety of groups (Zaldívar-Riverón et al. 2010, Hernández-Triana et al. 2012, Jalali et al. 2015). DNA barcoding is a rapid and efficient method to identify species based on the use of about 600 bp of the mitochondrial gene, cytochrome oxidase subunit 1 (Hebert et al. 2003).

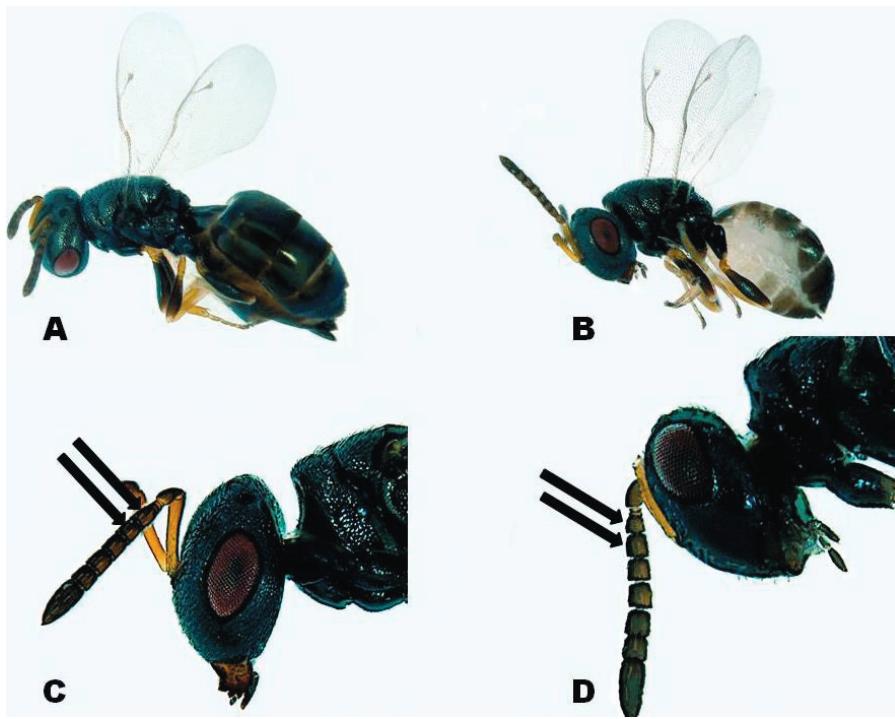


Fig. 2. Anatomical comparisons of sex differential characteristics of *A. calandrae*. A) Lateral habitus *A. calandrae* ♀, B) *A. calandrae* ♂, C) Female segments almost the same size, D) Male first funicular (third flagellar) segment shorter than the second.

Table 1. Nucleotide Sequence Changes in the CO1 Gene in *Anisopteromalus calandrae* from Veracruz, Mexico

Change position	MK272965	MG379893	AB690353	AB690354	Change type
175	A	G	A	A	*NoS (Met/Ile)
319	T	T	G	G	Synonymous
343	C	T	T	T	Synonymous
442	C	T	T	T	Synonymous
456	C	T	T	T	Synonymous
490	T	T	A	T	Synonymous
579	G	A	A	A	Synonymous

*NoS = Not synonymous

Of the species reported in the *Anisopteromalus* genus worldwide, *A. calandrae* is the only species in Mexico. Despite the importance of *A. calandrae* as a potential species for biological control against stored grain pests, no research has assessed whether it would be effective against *S. zeamais* in Mexico. In this study, we reported *A. calandrae* at Veracruz, Mexico, and corroborated identification by morphology and the barcode region (CO1). Sequences were generated to facilitate molecular diagnosis of the species in future studies.

References Cited

- Adarkwah, C., C. Ulrichs, S. Schaarschmidt, B. K. Badii, I. K. Addai, D. Obeng-Ofori, and M. Schöller. 2014. Potential of Hymenopteran larval and egg parasitoids to control stored-product beetle and moth infestation in jute bags. Bull. Entomol. Res. 104: 534-542.
- Baker, J. E., J. A. Fabrick, and K. Y. Zhu. 1998. Characterization of esterases in malathion-resistant and susceptible strains of the pteromalid parasitoid *Anisopteromalus calandrae*. Insect Biochem. Mol. Biol. 28: 1039-1050.
- Baur, H., Y. Kranz-Baltensperger, A. Cruaud, J. Y. Rasplus, A. V. Timokhov , and V. E. Gokhman. 2014. Morphometric analysis and taxonomic revision of *Anisopteromalus* Ruschka (Hymenoptera: Chalcidoidea: Pteromalidae) - an integrative approach. Syst. Entomol. 39: 691-709.
- Belda, C., and J. Riudavets. 2011. Reproduction of the parasitoids *Anisopteromalus calandrae* (Howard) and *Lariophagus distinguendus* (Förster) on arenas containing a mixed population of the coleopteran pests *Sitophilus oryzae* and *Rhyzopertha dominica*. J. Pest Sci. 85: 381-385.
- FAO, Organización de las Naciones Unidas para la Alimentación y la Agricultura. 2020. <http://www.fao.org/worldfoodsituation/csdb/es/> Accessed 25 November 2020.
- Folmer, O., M. Black, W. Hoeh, R. Lutz, and R. Vrijenhoek. 1994. DNA primers for amplification of mitochondrial Cytochrome C Oxidase Subunit I from diverse metazoan invertebrates. Mol. Marine Biol. Biotechnol. 3: 294-299.
- García-Lara, S., and D. J. Bergvinson. 2007. Integral Program to Reduce Post-harvest Losses in Maize. Agric. Téc. Méx. Vol.33 No.2 México may/ago 2007
- Ghimire, M. N., and T. W. Phillips. 2008. Effects of prior experience on host selection and host utilization by two populations of *Anisopteromalus calandrae* (Hymenoptera: Pteromalidae). Environ. Entomol. 37: 1300-1306.
- Giantsis, I. A., A. Chaskopoulou, and M. C. Bon. 2016. Mild-vectolysis: a nondestructive DNA extraction method for voucherizing sand flies and mosquitoes. J. Med. Entomol. 53: 692-695.
- Gupta, A., and P. M. Sureshan. 2014. A new pteromalid species of the genus *Anisopteromalus* Ruschka (Hymenoptera) from India. Orient. Insects 48: 67-72.
- Hebert, P. D. N., A. Cywinska, S. L. Ball, and J. R. DeWaard. 2003. Biological identifications through DNA barcodes. Proc. R. Soc. B: Biol. Sci. 270: 313-321.
- Hernández-Triana, L. M., J. L. Crainey, A. Hall, F. Fatih, J. Mackenzie-Dodds, A. J. Shelley, X. Zhou, R. J. Post, T. R. Gregory, and P. D. N. Hebert. 2012. DNA barcodes reveal cryptic genetic diversity within the blackfly subgenus *Trichodagmia* Enderlein (Diptera: Simuliidae: *Simulium*) and related taxa in the New World. Zootaxa 3514: 43-69.

- Jalali, S. K., R. Ojha, and T. Venkatesan. 2015. DNA barcoding for identification of agriculturally important insects, pp. 13-23. In *New Horizons in Insect Science: Towards Sustainable Pest Management*. Springer, New Delhi.
- Mahal, N., W. Islam, S. Parween, and K. A. M. S. H. Mondal. 2005. Effect of *Anisopteromalus calandrae* (Hymenoptera: Pteromalidae) in controlling residual populations of *Rhyzoperta dominica* (Coleoptera: Bostrichidae) in wheat stores. *Int. J. Trop. Insect Sci.* 25: 245-250.
- Noyes, J. S. 2022. Universal Chalcidoidea Database. World Wide Web electronic publication. <http://www.nhm.ac.uk/chalcidoids> Accessed 25 January 2022.
- Pingali, P. L., and S. Pandey. 2001. World maize needs meeting: technological opportunities and priorities for the public sector. In P. L. Pingali [ed.], CIMMYT 1999-2000. World maize facts and trends meeting world maize needs: Technological opportunities and priorities for the public sector. Centro Internacional para el Mejoramiento de Maíz y Trigo. El Batán, Estado de México, México.
- Pons, J., T. G. Barraclough, J. Gomez-Zurita, A. Cardoso, D. P. Duran, S. Hazell, and A. P. Vogler. 2006. Sequence-based species delimitation for the DNA taxonomy of undescribed insects. *Syst. Biol.* 55: 595-609.
- Schöller, M. E., P. W. Flinn, M. J. Grieshop, and E. Zd'a'rkova'. 2006. Biological control of stored product pests, pp. 67-87. In J. W. Heaps [ed.], *Insect Management for Food Storage and Processing*, 2nd ed. AACC International, Minnesota.
- SIAP (Servicio de Información Agroalimentaria y Pesquera). 2017. Anuario estadístico de la producción agrícola. SAGARPA. http://info.siap_siap.gob.mx:8080/agricola_siap_gobmx/AvanceNacionalSinPrograma.do Accessed 18 February 2017.
- Tigar, B. J., G. E. Key, M. E. Flores-S, and M. Vazquez-A. 1994. Field and post-maturity infestation of maize by stored product pest in Mexico. *J. Stored Prod. Res.* 30: 1-8.
- Zaldívar-Riverón, A., J. J. Martínez, F. S. Ceccarelli, V. S de Jesús-Bonilla, A. C Rodríguez-Pérez, A. Reséndiz-Flores, and M. A. Smith. 2010. DNA barcoding a highly diverse group of parasitoid wasps (Braconidae: Doryctinae) from a Mexican nature reserve. *Mitochondrial DNA* 21: 18-23.

Positive Edge Effects of Natural Habitats Produce Matching Response in Beetle Population and Plant Morphometry in Amaranth Crop

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Abstract. Components of a surrounding agricultural matrix can be important to the ecology of agroecosystems because they modulate biodiversity, abundance of pests (i.e., arthropods or weeds), and their natural enemies. Based on the assumption that organisms respond to edges, we assessed whether animals (beetles) and crop plants showed similar patterns across a gradient of perturbation and ecotones and natural habitats. Across the perturbation gradient, we analyzed (1) crop plant morphometry and (2) the structure of beetle populations in the perturbed area and natural habitats. Results showed that beetles (a bioindicator group) and *A. hypochondriacus* crop plants (perturbed area) responded similar to an ecotone, i.e., we found positive edge effects, but beetle richness negatively followed the gradient of perturbation affecting beetle nestedness and turnover. At the point nearest the ecotone, crop plants grew larger (e.g., panicle and plant height had the largest effects), which was consistent in time. Results supported the resource competition framework and resource distribution model in agroecosystems.

Introduction

Agroecological theory suggests natural adjacent vegetation reinforces microclimatic conditions in agroecosystems and provides food and shelter to wildlife that might increase ecosystem functions (Tscharntke et al. 2012, Clemente-Orta and Alvarez 2019). However, not all organisms respond the same to different types of vegetation, which frequently is related to the structure of the landscape and amount of habitat fragmentation (Ries and Sisk 2004; Laurance 2007; Tscharntke et al. 2012, 2016; Karp et al. 2018). A community in an ecosystem in transitional/colonization phase is different than a community in a well-established ecosystem (Prach and Walker 2011, Balmford et al. 2012). The rationale can be extrapolated to agroecosystems separating annual and perennial crops. Components of a surrounding agricultural matrix can be important to the ecology of agroecosystems because they modulate biodiversity, and therefore, abundance of pests (i.e., arthropods or weeds) and their natural enemies (Tscharntke et al. 2012, 2016; Sun

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et al. 2015; Karp et al. 2018; Clemente-Orta and Álvarez 2019; Álvarez 2021). For example, semi-natural habitats near or in orchards positively influence abundance of beneficial arthropods (and sometimes pests) (Alvarez et al. 2019a,b; Wan et al. 2019, 2021). However, because of intensive agricultural management, other crops near a growing field can be more important than semi-natural habitats to beneficial arthropods (Tscharntke et al. 2012, 2016; Clemente-Orta et al. 2020).

Grain amaranth, *Amaranthus hypochondriacus* L., is an emerging arable crop (Niveyro and Salvo 2014) cultivated worldwide (Caselato-Sousa and Amaya-Farfán 2012, Parra-Cota et al. 2014). Herbaceous plants are in the family Amaranthaceae in Caryophyllales and characterized by red to purple flowers in terminal or axial panicles (Alvarez et al. 2017). Amaranths are widely distributed in temperate zones (Mapes-Sánchez and Espitia-Rangel 2010, Das 2012). They are resistant to extreme weather and can grow in disturbed areas (Mapes-Sánchez and Espitia-Rangel 2010, Das 2012). Harvest of amaranth grain is an ancient practice (e.g., Mexico, Mapes-Sánchez and Espitia-Rangel 2010, Perez-Torres et al. 2011). An alternative way to obtain resources other than grain from the crop is to grow amaranth in the dry season and harvest leaves to use in meals.

Response of an amaranth crop and crop-related biodiversity to hedges and natural habitats has not been adequately studied in extreme environmental conditions of the dry season (Alvarez et al. 2016, 2017, 2019c). Beetles because of great diversity, relationship with plants and/or conservation status, and ecological specialization are used as bioindicator organisms to assess the status of ecosystems and agroecosystems (Spector 2006, Novelo et al. 2007, Syaripuddin et al. 2015). Based on the assumption that organisms respond to edges (Clemente-Orta and Álvarez 2019), we assessed whether animals and plants showed similar patterns across a gradient of perturbation. We analyzed plant morphometry in different zones in a perturbed area at different developmental times and the structure of the beetle population in the perturbation gradient. We hypothesized that natural vegetation would i) positively affect beetles following the gradient of perturbation, ii) have the same beetle species throughout the gradient, but iii) not affect plant growth.

Materials and Methods

The study was done in the dry season of 2015 at San Francisco Huilango (14°Q 54.58.78-20.82.669), Puebla, Mexico. San Francisco Huilango is part of Tochimilco municipality at 1,860 m above sea level and has 800 to 1,300 mm of annual precipitation (INEGI 2009, INAFED 2010). The region has large surfaces of natural and conserved forests near the Popocatepetl volcano (Fig. 1). Forty-three percent of the region is temperate forest, 3% deciduous forest, and 3% grassland.

We selected an area with different types of vegetation forming a perturbation gradient, i.e., deciduous forest, ecotone, and anthropized section that in this case was a crop of *A. hypochondriacus*. The remainder of the perimeter adjoined other crops, and there was no semi-natural vegetation between the areas.

Across the perturbation gradient three areas for comparison were delimited in a Z formation: 1) deciduous forest near the ecotone (natural area), (2) half of the growing area near the ecotone (hedge area), and (3) half of the growing area near adjoining growing areas (non-hedge area) (Fig. 1). In each area, we set three transects of 20 m parallel to the ecotone. Each transect was 10 m apart, and transects in areas were separated by a minimum of 100 m (Fig. 1C).¹

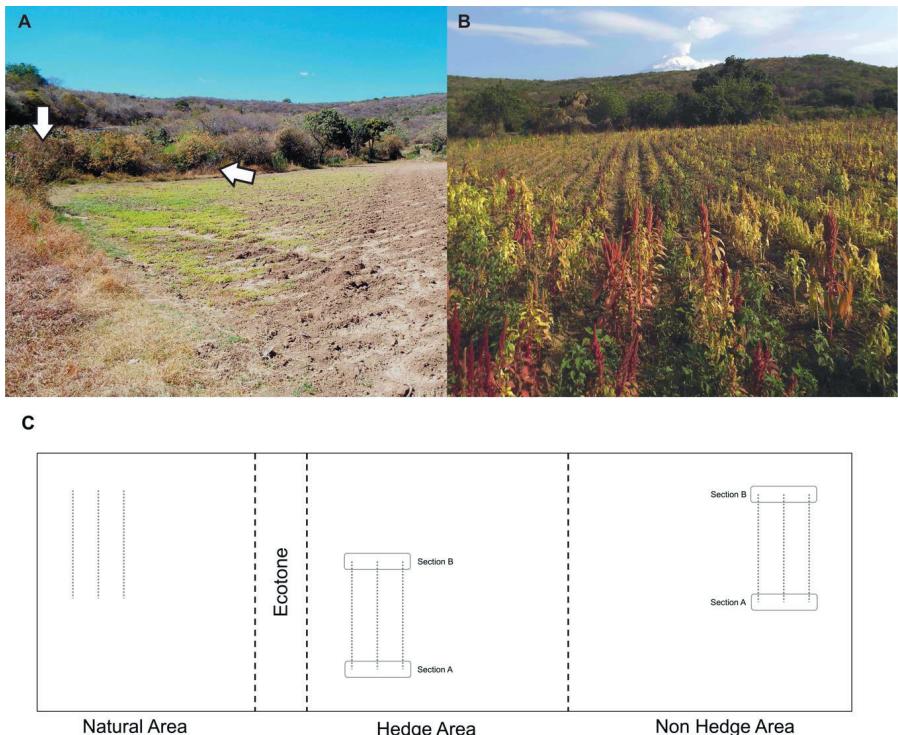


Fig. 1. Study area and sampling design. (A) Natural adjacent vegetation (white arrows) as an ecotone at the edge of the tilled growing area and next to the forest. (B) *Amaranthus hypocochndriacus* crop plants at the end of the dry season. (C) Form of the perturbation gradient and position of transects in each area.

Sixty *A. hypocochndriacus* plants were randomly selected at each of the hedge area and the non-hedge area, i.e., only plants at distal sections of each transect were used ($n = 10$). Section A was the south section of the transect, and Section B was the north section of the transect. Four morphometric variables of stalk width, leaf width, panicle length, and height of each plant were linearly measured with (1) vernier calipers (accuracy of 0.01 mm) and (2) a measuring tape (accuracy of 0.1 cm). The stalk was measured transversally at the middle point of the stalk. The leaf was measured transversally from edge to edge at the widest point of the leaf. Panicle was measured from the base of the panicle to the distal point. Plant height was measured from the base of the stalk to the apical point of the panicle. Measurements were made 2 months after sowing (Time 1) and at 4 months after sowing (Time 2).

Methods by Morón and Terron (2011) were used to sample plant foliage from April to July (once a month) in transects in the three areas. Beetles on foliage were collected by using an entomological net to sweep five randomly distributed plants. Samples were collected between 0900 and 1300 hours. Specimens were preserved in 70% alcohol and transported to a laboratory for identification. All specimens were identified to species and deposited in the entomological collection of the NGO, Institute of Research in Natural Sciences and Humanities (IINCINH).

Data on morphometric measurements of *A. hypochondriacus* were not distributed normally (Shapiro-Wilk normality test: stalk, $W = 0.877, p = 0.001$; leaf, $W = 0.942, p = 0.001$; panicle, $W = 0.829, p = 0.001$; height, $W = 0.909, p = 0.001$). Thus, correlation among all morphometric variables was examined by calculating pairwise Pearson's product-moment correlation coefficients. Generalized linear models (GLM) were fitted for each of the four variables. Each measured variable was used as the dependent variable, and time, area, and section as factors. Interactions among factors were analyzed.

Shannon and Simpson diversity indices on beetle richness and abundance calculated for each area were used to assess the structure of the beetle population among the study areas. Species accumulation curves and non-parametric estimators were calculated for the three areas together. Sampling effort for each area was calculated. Analyses were computed using EstimateS, v 9.1.0 software and SPADE (2009 actualization). Also analyzed was replacement in the form of richness gradients. The index of multiple-site dissimilarities (M_{sim}) of Baselga et al. (2007) was calculated by using:

$$M_{sim} = \frac{\sum_i S_i - S_T}{[\sum_{i < j} \min b_{ij}, b_{ji}] + [\sum_i S_i - S_T]}$$

where S_i is the richness at site i (α), S_T is the richness of the total of sites (γ), b_{ij} indicates the number of species that only occurred at site i but not site j , b_{ji} indicates the number of species that occurred only at site j but not site i , and min is the minimum number between b_{ij} and b_{ji} .

The index of multiple-site dissimilarities provides much help and confidence, because species replacement/turnover and loss could be the cause for differences in species composition among sites in a richness gradient. The index only responds to species replacement and does not depend on differences in species richness among sites, which is an advantage over other indices (Baselga 2007; Baselga et al. 2007, 2010, 2012). Function "beta.multi" from package "betapart" (Baselga et al. 2015) was used to compute the index of multiple-site dissimilarities. The function provides a list of values of similarity for (1) components of the replacement measured as Simpson's similarity, (2) nesting components measured as the nesting fraction resulting from Sorensen's similarity, and (3) overall (beta) diversity measured as Sorensen's similarity. Possible values are zero to one, where one indicated the maximum possible value (Baselga et al. 2015).

R v. 3.3.1 software was used to compute correlation analysis and the index of multiple-site dissimilarities. SPSS, v.19 software was used to compute GLM.

Results

Correlation analysis showed all variables were positively correlated. Height was the variable with the greatest correlation coefficients of all the variables (Table 1). Height and panicle had the greatest positive correlation coefficients, with exception of hedge area at Time 1. Conversely, stalk had the least correlation coefficients.

Table 1. Pairwise Pearson's Product Moment Correlations of Morphometric Variables of *Amaranthus hypochondriacus* Crop Plants (Hedge and Non-hedge Areas). Above Diagonal, *P*-values. Below Diagonal, Standard Correlation Coefficients.

		Time 1				Time 2			
		Hedge area		Non-hedge area		Hedge area		Non-hedge area	
		Stalk	Leaf	Panicle	Height	Stalk	Leaf	Panicle	Height
Stalk	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
Leaf	0.88	0.001	0.001	0.46	0.001	0.001	0.65	0.001	0.72
Panicle	0.48	0.42	0.001	0.48	0.78	0.001	0.53	0.46	0.001
Height	0.77	0.77	0.52	0.51	0.83	0.93	0.63	0.68	0.87

Table 2. Statistics from GLMs comparing Morphometric Variables of *Amaranthus hypochondriacus* Crop Plants, Comparing Time, Type (Hedge and Non-hedge Areas), Sections, and their Interactions.

Factor	n	Stalk			Leaf			Panicle			Height		
		Mean	SD	F	p	Mean	SD	F	p	Mean	SD	F	p
TIME	120	4.34	1.70	21.89	0.001	28.11	9.73	5.46	0.019	114.85	121.01	32.82	0.001
1	120	5.51	2.39	25.18	12.18					201.70	124.01		
2	120											454.00	224.82
TYPE	120	5.65	2.32	33.05	0.001	31.08	11.86	46.15	0.001	195.47	156.46	24.50	0.001
Hedge	120	4.20	1.69	22.21	8.18					121.08	80.97		
Non hedge	120											302.33	125.72
SECTION	120	5.34	2.37	11.54	0.001	29.49	12.68	20.12	0.001	183.52	136.48	11.59	0.001
A	120	4.50	1.82	23.80	8.38					133.04	117.96		
B	120											315.50	153.25
Interactions:													
TIME/TYPE	60	4.80	1.75	5.04	0.025	30.67	11.25	10.06	0.002	130.91	156.77	8.93	0.003
1/Hedge	60	3.88	1.53	25.54	7.13					98.80	66.58		
1/Non-hedge	60	5.84	2.61	31.49	12.53					260.03	127.74		
2/Hedge	60	6.50	2.51	18.88	7.84					143.37	88.22		
2/Non-hedge	60	4.52	1.79									333.15	130.23
TIME/SECTION	60	4.84	2.00	0.51	0.472	30.67	11.21	0.23	0.628	120.36	108.66	7.8	0.005
1/A	60	3.84	1.15	25.54	7.20					109.35	132.91		
1/B	60	5.84	2.61	28.31	13.99					246.67	132.86		
2/A	60	5.17	2.10	22.05	9.15					156.73	96.20		
2/B	60											377.18	155.77
TYPE/SECTION	60	6.54	2.38	15.75	0.001	36.68	12.22	21.27	0.001	236.55	150.56	5.05	0.025
Hedge/A	60	4.76	1.88	25.48	8.42					154.40	152.56		
Hedge/B	60	4.15	1.66	22.31	8.35					130.49	95.83		
Non-hedge/A	60	4.25	1.73	22.11	8.07					111.68	62.14		
Non-hedge/B	60											295.35	106.89

Table 2 summarizes statistics of each morphometric variable based on experimental design and GLM. According to analyses, variables showed differences in each factor. Variables, except leaf, were larger at Time 2. Sizes were larger in the hedge area than the non-hedge area. Section A had larger sizes than did Section B. There were interactions among all variables, i.e., between factors time and type, and between factors type and section. The interaction time-section was significant for the variable panicle. This indicated panicle was large in Section A, but larger at Time 2; i.e., it grew exponentially and doubled in size. Interaction type-section showed Section A in the hedge area had larger sizes. Conversely, other sections were similar with small sizes. Overall, patterns were stronger in panicle and height (Fig. 2).

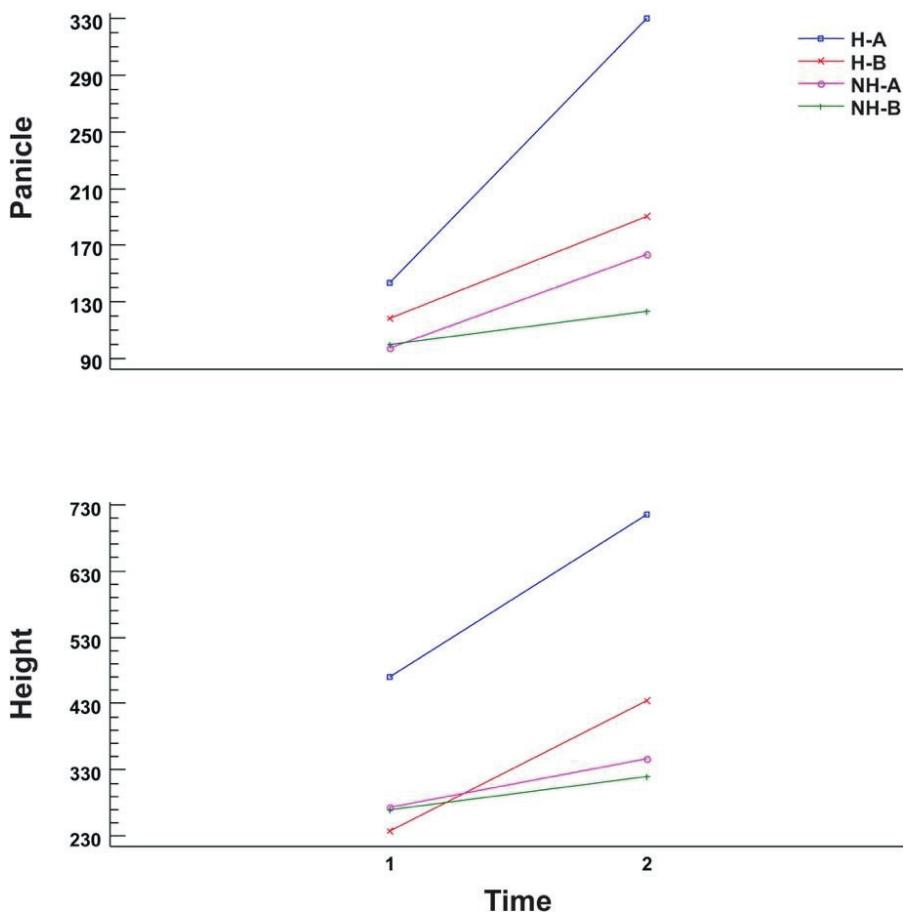


Fig. 2. Morphometric variables of *Amaranthus hypochondriacus* crop plants through time (Panicle and Height, scale in centimeters). Differences among transect sections (A or B) in the two areas in the agroecosystem, Hedge (H) and Non-hedge (NH) areas.

Collected were 21 beetle species in the families Brentidae, Chrysomelidae, Coccinellidae, Melyridae, Phengodidae, and Scarabaeidae. Table 3 shows relative abundance of beetle species, taxonomic information, and occurrence in different study areas. The family Chrysomelidae had most richness with 12 species, followed by the family Brentidae with three species. Coccinellidae and Melyridae each had two species, and the families Phengodidae and Scarabaeidae each had one species. *Diabrotica balteata* LeConte, 1865 was the most abundant species, followed by *Zygogramma signatipennis* (Stål, 1859), *Disonycha melanocephala* Jacoby, 1884, and *Celocephalapion buchanani* (Kissinger, 1958).

Fig. 3 shows the calculated species accumulation curve for overall data with 95% upper and lower values. Non-parametric estimators ACE, Chao 1, Chao 2, Jackknife 1, Jackknife 2, and ICE were visualized using data obtained in EstimateS software. ACE and Chao 1 were non-parametric estimators based on abundance and better adjusted to the asymptote. Specifically, Chao 1 is based on the number of rare species (singles and doubles) but in ACE, rare species are defined by the maximum number in abundance.

Sampling effort based on Chao 1 and ACE was 80% for hedge, 80% for non-hedge, and 60% for natural areas. Shannon (H') and Simpson (D) indices were high for the natural area ($H' = 2.370$, $D = 0.10494$), followed by the hedge area ($H' = 2.013$, $D = 0.17202$), and least for the non-hedge area ($H' = 0.955$, $D = 0.48097$).

The index of multiple-site dissimilarities showed great similarity among sites (components of the replacement value = 0.705), but no nestedness among sites (nesting component value = 0.094). The beetle community in the gradient was diversified (overall beta value = 0.8) (Fig. 4).

Discussion

Edge effects are changes in biotic or physical variables at transitions between adjoining habitats (Laurance 2007, Lopez-Barrera et al. 2007, Clemente-Orta and Álvarez 2019). Beetles (a bioindicator group) and *A. hypochondriacus* crop plants responded positively to an ecotone, which can be interpreted as positive edge effects. According to development of the experiment, *A. hypochondriacus* traits in Section A of the hedge area, the point nearest the ecotone, were larger. The pattern was consistent in time, i.e., panicle and plant height had the largest effects (Fig. 2). However, plant traits in Section B of the hedge area were more similar than in non-hedge areas. This showed the edge affected only plants nearest natural vegetation. Plant growth depends on conditions in a growing area. Recent accounts suggest that high-diversity plant assemblages in some scenarios benefit native plant-invaders rather than alien invaders, because of biogeographical history of plants species (Sun et al. 2015). If we accept that native plant species have better opportunities to settle when invading a new territory such as an agroecosystem, so species benefit by being near a native ecotone or hedge, we could explain the case for *A. hypochondriacus*. This amaranth species is native in Mesoamerican zones and has been cultivated for centuries in the study region (Mapes-Sánchez and Espitia-Rangel 2010).

Results showed low calculated sampling efficiency in the natural system; 12 species were collected in the area. This could be a result of several singletons in the final period of the sampling season which was the beginning of the rainy season. Beetle adults in the area usually emerge in May or June (M. A. Morón personal communication). It is possible that adults collected at the end of the sampling season belong to the beetle fauna of the rainy season.

Table 3. Taxonomic Information and Abundance of Beetle Species Associated with *Amaranthus hypochondriacus* in Natural (Deciduous Forest), Hedge, and Non-hedge Areas

Species	Family	Subfamily	Occurrence	Feeding type	Relative abundance (%)
<i>Acanthoscelides longistilus</i>	Chrysomelidae	Bruchinae	Natural, Hedge	Phytophagous	5.00
<i>Acanthoscelides oblectus</i>	Chrysomelidae	Bruchinae	Natural	Phytophagous	1.67
<i>Acanthoscelides pectoralis</i>	Chrysomelidae	Bruchinae	Natural	Phytophagous	1.67
<i>Acanthoscelides sp1</i>	Chrysomelidae	Bruchinae	Hedge	Phytophagous	1.67
<i>Acanthoscelides sp2</i>	Chrysomelidae	Bruchinae	Hedge	Phytophagous	1.67
<i>Attalus trimaculatus</i>	Melyridae	Malachiinae	No hedge	Phytophagous	1.67
<i>Coeloccephalapion buchananii</i>	Brentidae	Apioninae	Natural, Hedge	Phytophagous	6.67
<i>Coeloccephalapion sp1</i>	Brentidae	Apioninae	Hedge	Phytophagous	1.67
<i>Coeloccephalapion sp2</i>	Brentidae	Eumolpinae	Natural	Phytophagous	1.67
<i>Chalcophana sp</i>	Chrysomelidae	Melyridae	Natural	Phytophagous	5.00
<i>Colllops quadrimaculatus</i>	Coccinellidae	Malachiinae	Hedge	Predator	3.33
<i>Cryptognatha auriculata</i>	Coccinellidae	Scymninae	Natural, No hedge	Predator	3.33
<i>Diabrotica balteata</i>	Chrysomelidae	Galerucinae	Natural, Hedge, No hedge	Phytophagous	36.67
<i>Disonycha melanocephala</i>	Chrysomelidae	Galerucinae	No hedge	Phytophagous	6.67
<i>Disonycha sp1</i>	Chrysomelidae	Galerucinae	Hedge	Phytophagous	1.67
<i>Euphoria indica</i>	Scarabaeidae	Cetoniinae	Natural	Phytophagous	3.33
<i>Lachnaia sp</i>	Chrysomelidae	Cryptocephalinae	Natural	Phytophagous	1.67
<i>Phengodes sp</i>	Phengodidae	-	Natural	Predator	1.67
<i>Platothispa gregorii</i>	Chrysomelidae	Hispinae	Natural	Phytophagous	1.67
<i>Scymnus (Pullus) loewii</i>	Coccinellidae	Scymninae	Hedge	Predator	3.33
<i>Zygogramma signatipennis</i>	Chrysomelidae	Chrysomelinae	Hedge	Phytophagous	8.33

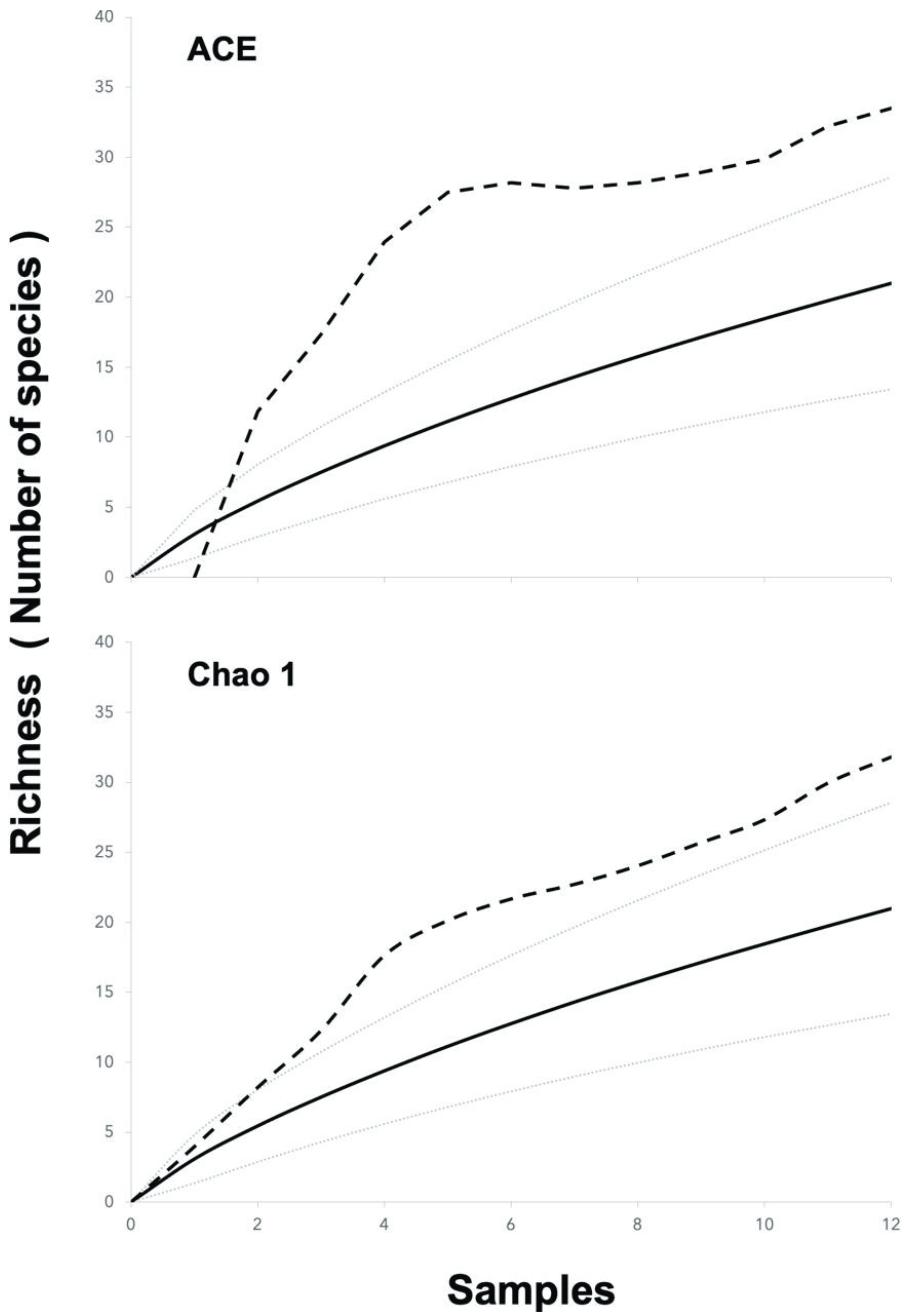


Fig. 3. Accumulation curves of beetle richness with ACE and Chao1. Species accumulation (solid line) with 95% CI (dashed gray) and estimator (dashed black).

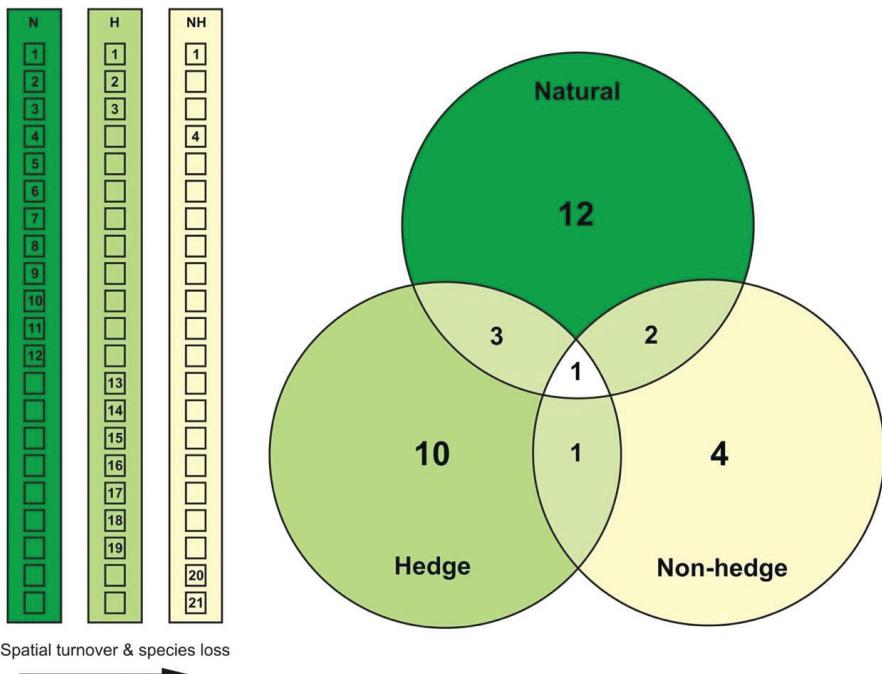


Fig. 4. Nestedness and turnover of beetle population associated with *Amaranthus hypocondriacus* in Natural (Deciduous Forest, N), Hedge (H), and Non-hedge (NH) areas. Rectangles represent spatial turnover and species loss. Numbers inside squares represent beetle species from most to least common and are separated by abundance. Circles represent the ecological community showing the number of species in each study area and shared between areas.

Beetles negatively followed the perturbation gradient, i.e., gradient related with quality of habitat (ecological habitats, Colwell et al. 2004, Clemente-Orta and Álvarez 2019). The natural area (deciduous forest) was most diverse, followed by the hedge area, and the non-hedge area. Species richness had transitional response, i.e., richness responded to natural adjacent vegetation in negative form in the natural system (high-quality habitat) and in a positive form in the crop (low-quality habitat). The species were not nested, therefore, communities with low richness were not subsets of communities with high richness (Fig. 4). Only four species were shared among areas, especially the natural area, *A. longistilus*, *C. buchanani*, *Cryptognatha auriculata* Mulsant, 1850, and *D. balteata* (Table 3). Almost all species were phytophagous except four predators: *C. auriculata*, *Scymnus (Pullus) loewii* Mulsant, 1850, *Collops quadrimaculatus* (Fabricius, 1798), and *Phengodes* sp. The four species were only in the growing area, especially in the hedge area (but *C. auriculata* was shared between the natural area and non-hedge area Fig. 4), which shows the need for prey and support edge effects in the crop. This was probably related to vigorous *A. hypocondriacus* plants and microclimatic features of the ecotone, closely relating the response of diversity with plant morphology.

Overall, our three study areas form a matrix of very different properties. According to classic rationale by Duelli et al. (1990), the growing area in the study had properties of a hard edge, but the ecotone had properties of a soft edge. Thus, we explain positive edge effects in plant growth and beetles in context of a resource competition framework (Sun et al. 2015) and/or a resource distribution model (Ries and Sisk 2004, Macreadie et al. 2010). Complementary resource distribution refers to a scenario where two adjacent habitats have different resource availability (quantity and quality). When the habitats are very different, a complementary distribution of resources will drive a positive response, i.e., low-quality habitat will have less abundance of animals than other habitats, but individuals at the boundaries will benefit from availability of new resources. The resource-based model shows resources could be concentrated at an edge, therefore when natural habitats are fragmented, resources are better exploited around edges by native and/or opportunistic species (Ries and Sisk 2004, Macreadie et al. 2010), which could be the case of a *A. hypochondriacus* crop.

The study is the first to understand interactions among grain amaranth agroecosystems, biodiversity, and natural or semi-natural habitats. Only our first hypothesis was true. The gradient of perturbation and the natural habitat affected beetle nestedness and turnover and growth of amaranth plants. The specific pattern on a local scale (small-holder) suggested landscape-scale perspective is needed in future research to prove if response to proximity to natural adjacent vegetation, shown by *A. hypochondriacus*, is a rule in dry-season agroecosystems.

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References Cited

- Álvarez, H. A. 2021. Semi-natural habitats and natural enemies in olive orchards: abundance, function, trophic interactions, and global climate change. University of Granada, Spain. ISBN: 9788411170376.
- Álvarez, H. A., H. Carrillo-Ruiz, and M. A. Morón. 2016. Record of Scarabaeoidea larvae and adults associated with *Amaranthus hypochondriacus* L. and living fences. Southwest. Entomol. 41: 675-679.
- Álvarez, H. A., H. Carrillo-Ruiz, D. Jiménez-García, and M. A. Morón. 2017. Abundance of insect fauna associated with *Amaranthus hypochondriacus* L. crop, in relation to natural living fences. Southwest. Entomol. 42: 131-135.
- Álvarez, H. A., M. Morente, F. S. Oi, E. Rodríguez, M. Campos, and F. Ruano. 2019a. Semi-natural habitat complexity affects abundance and movement of natural enemies in organic olive orchards. Agric. Ecosyst. Environ. 285: 106618.
- Álvarez, H. A., M. Morente, M. Campos, y F. Ruano. 2019b. La madurez de las cubiertas vegetales aumenta la presencia de enemigos naturales y la resiliencia de la red trófica de la copa del olivo. Ecosistemas 28: 92-106.
- Álvarez, H. A., H. Carrillo-Ruiz, J. F. López-Olguín, D. Jiménez-García, and M. A. Morón. 2019c. Beetles in *Amaranthus hypochondriacus* L. during the dry season. Southwest. Entomol. 44: 199-204.

- Álvarez, H. A., M. Morente, M. Campos, and F. Ruano. 2021. Ground cover presence in organic olive orchards affects the interaction of natural enemies against *Prays oleae*, promoting an effective egg predation. Agric. Ecosyst. Environ. 107441.
- Balmford, A., R. Green, and B. Phalan. 2012. What conservationists need to know about farming. P. Roy. Soc. B-Biol. Sci. rspb: 20120515.
- Baselga, A. 2007. Disentangling distance decay of similarity from richness gradients: response to Soininen et al. 2007. Ecography 30: 838-841.
- Baselga, A. 2010. Partitioning the turnover and nestedness components of beta diversity. Glob. Ecol. Biogeogr. 19: 134-143.
- Baselga, A. 2012. The relationship between species replacement, dissimilarity derived from nestedness, and nestedness. Glob. Ecol. Biogeogr. 21: 1223-1232.
- Baselga, A., A. Jiménez-Valverde, and G. Niccolini. 2007. A multiple-site similarity measure independent of richness. Biol. Letters 3: 642-645.
- Baselga, A., D. Orme, S. Villéger, J. De Bortoli, and F. Leprieur. 2015. Partitioning beta diversity into turnover and nestedness components. <https://cran.r-project.org/web/packages/betapart/betapart.pdf> Accessed 20 September 2016.
- Caselato-Sousa, V. M., and J. Amaya-Farfán. 2012. State of knowledge on amaranth grain: a comprehensive review. J. Food Sci. 77: 93-104.
- Clemente-Orta, G., y H. A. Alvarez. 2019. La influencia del paisaje agrícola en el control biológico desde una perspectiva espacial. Ecosistemas 28: 13-25.
- Clemente-Orta, G., F. Madeira, I. Batuecas, S. Sossai, A. Juárez-Escario, and R. Albajes. 2020. Changes in landscape composition influence the abundance of insects on maize: the role of fruit orchards and alfalfa crops. Agric. Ecosyst. Environ. 291: 106805.
- Colwell, R. K., C. Rahbek, and N. J. Gotelli. 2004. The Mid-Domain Effect and species richness patterns: what have we learned so far? Am Nat. 163: E1-E23.
- Das, S. 2012. Systematics and taxonomic delimitation of vegetable, grain and weed amaranths: a morphological and biochemical approach. Gen. Res. Crop Evol. 59: 289-303.
- Duelli, P., M. Studer, I. Marchand, and S. Jakob. 1990. Population movements of arthropods between natural and cultivated areas. Biol. Conserv. 54: 193-207.
- INAFED (Instituto Nacional para el Federalismo y el Desarrollo Municipal). 2010. Enciclopedia de los Municipios. http://www.elocal.gob.mx/wb/ELOCAL/ELOC_Enciclopedia Accessed 20 September 2016.
- INEGI (Instituto Nacional de Estadística, Geografía e Informática). 2009. Prontuario de información geográfica municipal de los Estados Unidos Mexicanos 21188.
- Karp, D. S., R. Chaplin-Kramer, T. D. Meehan, E. A. Martin, F. DeClerck, et al. 2018. Crop pests and predators exhibit inconsistent responses to surrounding landscape composition. Proc. Nat. Acad. Sci. 115: E7863-E7870.
- Laurance, W. F. 2007. Ecosystem decay of Amazonian forest fragments: implications for conservation, pp. 9-35. In T. Tscharntke, C. Leuschner, M. Zeller, E. Guhardja, and A. Bidin [eds.], Stability of Tropical Rainforest Margins. Springer, Berlin, Germany.
- López-Barrera F., J. J. Armesto, G. Williams-Linera, C. Smith-Ramírez, and R. H. Manson. 2007. Fragmentation and edge effects on plant-animal interactions, ecological processes and biodiversity, pp. 69-101. In A. C. Newton [ed.],

- Biodiversity Loss and Conservation in Fragmented Forest Landscapes. The Forests of Montane Mexico and Temperate South America. CABI, Oxfordshire, UK.
- Macreadie, P. I., J. S. Hindell, M. J. Keough, G. P. Jenkins, and R. M. Connolly. 2010. Resource distribution influences positive edge effects in a seagrass fish. *Ecology* 91: 2013-2021.
- Mapes-Sánchez, E. C., y E. Espitia-Rangel. 2010. Recopilación y análisis de la información existente de las especies del género *Amaranthus* cultivadas y de sus posibles parientes silvestres en México. CONABIO, Mexico.
- Morón, M. A., y R. A. Terrón. 2011. Entomología práctica. Una guía para el estudio de los insectos con importancia agropecuaria, médica, forestal y ecológica de México. Instituto de Ecología A.C., Xalapa, México. Edición electrónica 2011.
- Niveyro, S., and A. Salvo. 2014. Taxonomic and functional structure of phytophagous insect communities associated with grain amaranth. *Neotropical Entomology* 43: 532-540.
- Novelo, E. R., H. Delfin-Gonzales, and M. A. Morón. 2007. Copro-necrophagous beetle (Coleoptera: Scarabaeidae) diversity in an agroecosystem in Yucatan, Mexico. *Revista de Biología Tropical* 55: 83-99.
- Parra-Cota, F. I., J. J. Peña-Cabriales, S. de los Santos-Villalobos, N. A. Martínez-Gallardo, and J. P. Délano-Frier. 2014. *Burkholderia ambifaria* and *B. caribensis* promote growth and increase yield in grain amaranth (*Amaranthus cruentus* and *A. hypochondriacus*) by improving plant nitrogen uptake. *PloS One* 9: 88094.
- Pérez-Torres, B. C., A. Aragón-García, R. Pérez-Avilés, L. R. Hernández, and J. F. López-Olguín. 2011. Entomofaunistic study in of the cultivation of amaranth (*Amaranthus hypochondriacus* L.) in Puebla, México. *Rev. Mex. Cienc. Agric.* 2: 359-371.
- Prach, K., and L. R. Walker. 2011. Four opportunities for studies of ecological succession. *Trends in Ecology and Evolution* 26: 119-123.
- Ries, L., and T. D. Sisk. 2004. A predictive model of edge effects. *Ecology* 85: 2917-2926.
- Spector, S. 2006. Scarabaeine dung beetles (Coleoptera: Scarabaeidae: Scarabaeinae): an invertebrate focal taxon for biodiversity research and conservation. *Coleopterists Society Monographs: Patricia Vaurie Series* 5: 71-83.
- Sun, Y., H. Müller-Schärer, J. L. Maron, and U. Schaffner. 2015. Origin matters: diversity affects the performance of alien invasive species but not of native species. *Am. Nat.* 185: 725-736.
- Syaripuddin, K., K. W. Sing, and J. J. Wilson. 2015. Comparison of butterflies, bats and beetles as bioindicators based on four key criteria and DNA barcodes. *Tropical Conservation Science* 8: 138-149.
- Tscharntke, T., J. M. Tylianakis, T. A. Rand, et al. 2012. Landscape moderation of biodiversity patterns and processes - eight hypotheses. *Biol Rev.* 87: 661-685.
- Tscharntke, T., D. S. Karp, R. Chaplin-Kramer, et al. 2016. When natural habitat fails to enhance biological pest control - five hypotheses. *Biol. Control* 204: 449-458.
- Wan, N. F., X. Y. Ji, J. Y. Deng, L. P. Kiær, Y. M. Cai, and J. X. Jiang. 2019. Plant diversification promotes biocontrol services in peach orchards by shaping the ecological niches of insect herbivores and their natural enemies. *Ecol Indic.* 99: 387-392.

New Records of Weevils¹ for the State of Durango and Mexico

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Abstract. Seventeen species of curculionids were reported for the first time for the State of Durango, of which five were new records for Mexico: *Centrinites strigicollis* Casey, 1892; *Smicronyx vestitus* LeConte, 1876; *Smicronyx tardus* Dietz, 1894; *Sibinia texana* (Pierce, 1908); and *Chalcodermus martini* Van Dyke, 1930. Species in the subfamily Baridinae were the most abundant in the study (43.5%), with four tribes, six genera, and 10 species.

Resumen. Se reportan por primera vez 17 especies de curculiónidos para el Estado de Durango; cinco de estas son nuevos registros para México: *Centrinites strigicollis* Casey, 1892; *Smicronyx vestitus* LeConte, 1876; *Smicronyx tardus* Dietz, 1894; *Sibinia texana* (Pierce, 1908); y *Chalcodermus martini* Van Dyke, 1930. La subfamilia Baridinae fue la más abundante (43.5%) con cuatro tribus, seis géneros, y 10 especies.

Introduction

Although Durango is the fourth largest state in Mexico, invertebrate fauna there has not been well studied. Studies of weevils (Curculionidae) have been sporadic and directed at pest species such as *Anthonomus grandis* Boheman, 1843, collected in pheromone traps near the Municipality of Tlahualilo, Durango (Kim et al. 2006) and *Scyphophorus acupunctatus* Gyllenhal, 1838 and *Peltophorus polymitus* Boheman, 1845 collected from *Agave durangensis* Gentry, 1982 (Reyes et al. 2021). This study was part of the "Entomofauna de Picudos de México" research program of the Universidad Autónoma Agraria Antonio Narro-Buenavista (UAAAN-B), by the Buenavista-Department of Agricultural Parasitology (DPA-B), whose main objective is contributing to knowledge of Curculionidae in Mexico. Here are results for the State of Durango.

Materials and Methods

The study was done in the State of Durango, Mexico between July 2020 and August 2021. Adult weevils were collected with an entomological net, 100 m from any road or path, among undergrowth, shrubs, trees, and crops apple (*Malus domestica* Borkh), alfalfa (*Medicago sativa* L.), and maize (*Zea mays* L.). Adult

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specimens collected were put into previously labeled bottles, filled with 70% ethyl alcohol, and transferred to the Insect and Mite Taxonomy Laboratory of the DPA-B at UAAAN-B. They were identified by using taxonomic keys by Dietz (1896), Champion (1902-1909), Sleeper (1957), Burke (1960), Anderson (1962), Clark (1978), Clark and Burke (2005), Prena (2009), and Soto (2017). The Taxonomic Catalog of Species of Mexico (Ordoñez et al. 2008) and the Global Biodiversity Information Infrastructure (GBIF 2022) were consulted. The taxonomic classification used was that proposed by Anderson (2002). The identified specimens were deposited in the DPA-B at UAAAN-B Insect Collection, and other specimens are in the Zaragoza Experimental Site Insect Collection CIRNE-INIFAP at Zaragoza, Coahuila.

Results and Discussion

Sixty-two adults Curculionidae were captured from seven subfamilies, 13 tribes, and 26 species, with Baridinae the most abundant subfamily (43.5%) with four tribes, six genera, and 10 species. Entiminae was 35.4%, with two tribes, three genera, and five species. Curculioninae was 12.9% with four tribes, four genera, and seven species. Among other subfamilies were one tribe, one genera, and one species, respectively. *Linogeraeus hospes* Kuschel, 1983 was the most represented species (19.3%) of all the specimens. Seventeen new records were determined for the State of Durango, of which five were new records for Mexico. Species of Curculionidae collected in the State of Durango were:

Subfamily Curculioninae Latreille, 1802.

Tribe Anthonomini Thomson, 1859.

Anthonomus stolatus Fall, 1901. One specimen. Mexico, Durango, Canatlán Km. 48. 24°25'28"N, 104°41'55"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Southern United States of America; in Mexico, in the State of Baja California Norte (Clark and Burke 2005, GBIF 2022). New record for the State of Durango.

Tribe Smicronychini Seidlitz, 1891.

Smicronyx spretus Dietz, 1894. One specimen. Mexico, Durango, San Lucas de Ocampo Km. 54. 24°28'14"N, 104°41'44"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Southwestern United States of America; in Mexico, in the States of Baja California North and South, Nuevo León, Sonora, and Tamaulipas (Anderson 1962, GBIF 2022). New record for the State of Durango.

Smicronyx tardus Dietz, 1894. One specimen. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Western United States of America (Anderson 1962, GBIF 2022). For Mexico, it was the first record from the State of Durango.

Smicronyx vestitus LeConte, 1876. One specimen. Mexico, Durango, Ocampo Km. 307. 26°18'08"N, 105°10'17"W. 1/VIII/2020. Collector Y. Juarez-Ortiz.

Distribution: Eastern United States of America (Anderson 1962, GBIF 2022). For Mexico, it was the first record from the State of Durango.

Tribe Tychiini Thomson, 1859.

Sibinia texana (Pierce, 1908). One specimen. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Southern Texas, United States of America (Clark 1978). For Mexico, it was the first record from the State of Durango.

Sibinia triseriata Clark, 1978. One specimen. Mexico, Durango, Ocampo Km. 307. 26°18'08"N, 105°10'17"W. 1/VIII/2020. Collector Y. Juarez-Ortiz.

Distribution: Southeastern United States of America; in Mexico, in the States of Nuevo León and Tamaulipas. New record for the State of Durango (Clark 1978, GBIF 2022).

Incertae sedis

Macrorhoptrus sphaeralciae Pierce, 1908. Two specimens. Mexico, Durango, Ocampo Km. 307. 26°18'08"N, 105°10'17"W. 1/VIII/2020; Km. 118 José María, 24°52'47"N, 104°27'18"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Southwestern United States of America; in Mexico, in the States of Querétaro (GBIF 2022, Jones and Luna-Cozar 2007). New record for the State of Durango.

Subfamily Baridinae Schoenherr, 1836.

Tribe Baridini Schoenherr, 1836.

Baris futilis Casey, 1892. Two specimens. Mexico, Durango, El Calabazal Km. 271. 24°00'52"N 104°29'21"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Southern United States of America; also in Mexico, in the State of Baja California (GBIF 2022). New record for the State of Durango.

Baris striata (Say, 1831). Four specimens. Mexico, Durango, Guadalupe Victoria Km. 40. 24°25'13"N, 104°11'25"W. 31/VII/2020; Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020; Ocampo Km. 307. 26°18'08"N, 105°10'17"W. 1/VIII/2020. Collector Y. Juarez-Ortiz.

Distribution: Central and Eastern United States of America; in Mexico, in the State of Baja California (GBIF 2022). New record for the State of Durango.

Trichobaris soror Champion, 1909. One specimen. Mexico, Durango, Rodeo Km. 154. 25°07'52"N, 104°32'27"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Mexico, in the states of Chihuahua, Guanajuato, Hidalgo, Mexico, Oaxaca, Puebla, and Querétaro (GBIF 2022, Jones and Luna-Cozar 2007). New record for the State of Durango.

Tribe Madarini Jekel, 1865.

Stictobaris ornatella Casey, 1920. Two specimens. Mexico, Durango, Guadalupe Victoria Km. 40. 24°25'13"N, 104°11'25"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Southern United States of America; in Mexico, in the State of Durango (Champion 1902-1906, GBIF 2022).

Tribe Madopterini Lacordaire, 1866.

Linogeraeus hospes (Casey, 1892). Twelve specimens. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020; El Calabazal Km. 271. 24°00'52"N, 104°29'21"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Arizona, New Mexico, and Texas in the southern United States of America; throughout Mexico and Central America (Prena 2009).

Linogeraeus tonsilis (Bohemian, 1844). One specimen. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: From southern United States of North America, throughout Mexico, to Costa Rica (Prena 2009).

Linogeraeus capillatus (LeConte, 1876). One specimen. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: From Central United States of America, throughout Mexico, to northern South America, and at La Española Island (Prena 2009).

Linogeraeus x-notatum Champion, 1908. One specimen. Mexico, Durango, La Joya Km 26. 24°13'26"N, 104°42'34" W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Mexico, in the States of Chiapas, Guerrero, and Tamaulipas (Champion 1902-1906, GBIF 2022). New record for the State of Durango.

Centrinites strigicollis Casey, 1892. Two specimens. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Eastern United States of America. New record for Mexico and for the State of Durango (Champion 1902-1906, GBIF 2022).

Nicentrites testaceipes (Champion, 1908). One specimen. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Mexico, in the States of Coahuila, Oaxaca, and Querétaro (GBIF 2022, Jones and Luna-Cozar 2007). New record for the State of Durango.

Subfamily Ceutorhynchinae Gistel, 1856.

Tribe Cnemogonini Colonnelli, 1979.

Auleutes asper (LeConte, 1876). One specimen. Mexico, Durango, Guadalupe Victoria Km. 40. 24°25'13"N 104°11'25"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Canada; United States of America; in Mexico, in the States of Baja California and Chihuahua. New record for the State of Durango (Dietz 1896, GBIF 2022).

Subfamily Cryptorhynchinae Schoenherr, 1825.

Tribe Cryptorhynchini Schoenherr, 1825.

Phydenus mucireus (Germar, 1823). One specimen. Mexico, Durango, Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: In Arizona in United States of America; in Mexico, in the States of Durango, Guerrero, Oaxaca, and Puebla; in Brazil, Costa Rica, Guatemala, and Nicaragua (Champion 1902-1906, GBIF 2022).

Subfamily Entiminae Schoenherr, 1823.

Tribe Naupactini Gistel, 1856.

Megalostylus splendidus Chevrolat, 1878. One specimen. Mexico, Durango, San Juan del Río Km. 118. 24°52'47"N, 104°27'18"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Mexico in the States of Durango, Guerrero, Mexico, Mexico City, Michoacán, Puebla, and Sonora (Champion 1902-1906, GBIF 2022).

Naupactus cervinus Boheman, 1840. One specimen. Mexico, Durango, San Lucas de Ocampo Km. 54. 24°28'14"N, 104°41'44"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: It essentially is found throughout the world (GBIF 2022).

Pantomorus albosignatus Boheman, 1840. One specimen. Mexico, Durango, Ocampo Km. 307. 26°18'08"N, 105°10'17"W. 1/VIII/2020. Collector Y. Juarez-Ortiz.

Distribution: United States of America; Guatemala; in Mexico, in the States of Aguascalientes, Chiapas, Chihuahua, Coahuila, Durango, Guanajuato, Guerrero, Jalisco, Mexico, Nuevo León, Oaxaca, Puebla, Querétaro, San Luis Potosí, Tamaulipas, and Zacatecas (GBIF 2022, Jones and Luna-Cozar 2007).

Tribe Sciaphilini Sharp, 1891.

Mitostylus setosus (Sharp, 1891). Ten specimens. Mexico, Durango, Guadalupe Victoria Km. 40. 24°25'13"N, 104°11'25"W. 31/VII/2020; Nombre de Dios Km. 225. 23°49'03"N, 104°08'26"W. 31/VII/2020; San Lucas de Ocampo Km. 54. 24°28'14"N, 104°41'44"W. 26/VIII/2021; El Calabazal Km. 271. 24°00'52"N, 104°29'21"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: Southern United States of America; in Mexico, in the States of Aguascalientes, Chihuahua, Coahuila, Colima, Durango, Guanajuato, Hidalgo, Jalisco, Mexico, Mexico, Nuevo León, Puebla, Querétaro, San Luis Potosí, Tabasco, Tamaulipas, Veracruz, and Zacatecas (GBIF 2022).

Mitostylus tenuis Horn, 1876. Nine specimens. Mexico, Durango, Ocampo Km. 307. 26°18'08"N, 105°10'17"W. 1/VIII/2020; Guadalupe Victoria Km. 40. 24°25'13"N, 104°11'25"W. 31/VII/2020; San Juan del Río Km. 118. 24°52'47"N, 104°27'18"W. 26/VIII/2021; Rodeo Km. 154, 25°07'52"N, 104°32'27"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: Central and Southern United States of America, in Mexico, in the State of Nuevo León (GBIF 2022). New record for the State of Durango.

Subfamily Lixinae Schoenherr, 1823.

Tribe Lixini Schoenherr, 1823.

Microlarinus lypriformis Wollaston, 1861. One specimen. Mexico, Durango, Rodeo Km. 154. 25°07'52"N, 104°32'27"W. 26/VIII/2021. Collector Y. Juarez-Ortiz.

Distribution: In Arizona and California in the United States of America; in Mexico, in the States of Baja California Sur, Chihuahua, Sonora, and Veracruz. New record for the State of Durango (Champion 1902-1906, GBIF 2022).

Subfamily Molytinae Schoenherr, 1823.

Tribe Sternechini Lacordaire, 1863.

Chalcodermus martini Van Dyke, 1930. Two specimens. Mexico, Durango, El Calabazal Km. 271. 24°00'52"N, 104°29'21"W. 31/VII/2020. Collector Y. Juarez-Ortiz.

Distribution: In Arizona in the United States of America. The first record from Mexico and for the State of Durango (Champion 1902-1906).

Identification of curculionids in Mexico is limited because of lack of taxonomic keys and a limited number of taxonomists, because most taxonomists are in other countries (Soto 2017). Systematics of the family at almost all levels is controversial; for example, *Macrorhoptus sphaeralciae* was not assigned any tribe, and *Linogeraeus* is a genus whose taxonomic location at tribe and species levels depends on the author; synonyms of species cannot be recorded with certainty to species as is the case for *Naupactus cervinus* = *Pantomorus cervinus* = *Asynonychus cervinus*; and *Pantomorus albosignatus* = *Naupactus albosignatus*.

Curculionids are widely distributed throughout the world, in almost all known habitats, terrestrial and aquatic. Information is lacking on distribution, ecology, natural history, and immature stages of most Mexican species (Soto 2017). This is mostly because sampling has been sporadic and aimed at species of economic importance. Some regions, especially the northern states of Mexico have no or few records including the State of Durango for which there were no reports of faunal studies, and therefore, a large number of new records was obtained, both for the state and for the country.

Of the species sampled, some of economic importance were identified, such as *Naupactus cervinus* that attacks foliage and roots of fruit, ornamental, and forest species (Rosas et al. 2011). In lemon trees, adults feed on immature shoots and

mature leaves (Sumano et al. 2014). *Stictobaris ornatella* bores into the stem of amaranth plants, which causes wilting and bending of the plant, as the larvae of such species bore through the stem from the neck to the base of the inflorescence (Salas and Boradonenko 2006). *Linogeraeus capillatus* became important in the maize agroecosystem, because it is commonly and more frequently found in the crop. Perez et al. (2011) reported that larvae stratify maize plants; most first-instar larvae are on leaf veins, while second and third instars are on the leaf sheath, and fourth and fifth instars are on nodes and the inner stem, which can significantly damage the plant.

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References Cited

- Anderson, D. M. 1962. The weevil genus *Smicronyx* in America, North of Mexico (Coleoptera: Curculionidae). Proc. U.S. Natl. Museum 113: 185-372.
- Anderson, R. S. 2002. Family 131. Curculionidae Latreille 1802, pp. 722-806. In R. H. Arnett, Jr., M. C. Thomas, P. E. Skelley, and F. J. Howard [eds.], American Beetles, Vol. II: Polyphaga: Scarabaeoidea through Curculionoidea. CRC Press LLC, Boca Raton, FL.
- Burke, H. R. 1960. A new genus and two new species of weevils from Texas with notes on others (Curculionidae). Coleopt. Bull. 14: 121-127.
- Champion, G. C. 1902-1906. Insecta. Coleoptera. Rhynchophora, Curculionidae, Curculioninae (part), pp. 1-713. In R. H. Porter [ed.], Biología Centrali-Americana. Vol. 4. Part 4. London.
- Clark, W. K. 1978. The weevil genus, *Sibinia* Germar: natural history, taxonomy, phylogeny, and zoogeography, with revision of the New World species (Coleoptera: Curculionidae). Quaest. Ent. 14: 91-387.
- Clark, W. E., and H. R. Burke. 2005. Revision of the subgenus *Cnemocyllus* Dietz of the weevil genus *Anthonomus* Germar (Coleoptera: Curculionidae, Anthonomini). Insecta Mundi 19: 1-54.
- Dietz, W. G. 1896. Revision of the Genera and Species of Ceutorhynchini Inhabiting North America. Trans. Am. Entomol. Soc. 23: 387-480.
- GBIF Secretariat. 2022. GBIF Backbone Taxonomy. Checklist dataset <https://doi.org/10.15468/39omei> Accessed via GBIF.org on 15 February 2022.
- Jones, R. W., and J. Luna-Cozar. 2007. Lista de las especies de Curculionoidea (Insecta: Coleoptera) del estado de Querétaro, México. Acta Zool. Mex. 23: 59-77.
- Kim, K. S., P. Cano-Ríos, and T. W. Sappington. 2006. Using genetic markers and population assignment techniques to infer origin of boll weevils (Coleoptera: Curculionidae) unexpectedly captured near an eradication zone in Mexico. Environ. Entomol. 35: 813-826.
- Ordóñez, M. M., R. Muñiz, and F. Gama. 2008. Catálogo taxonómico de especies de México, pp. 5-338. In J. Soberón, G. Halfitter, y J. Llorente-Bousquets [eds.], Capital Natural de México, Vol. I: Conocimiento Actual de la Biodiversidad. Comisión Nacional para el Conocimiento y Uso de la Biodiversidad

- (CONABIO), México. CD1. http://www2.biodiversidad.gob.mx/pais/pdf/CapNatMex/Vol%201/I00_PrefacioGuia.pdf
- Perez, K., N. Villarreal, and C. Fernández. 2011. Bioecología del picudo del tallo del maíz *Linogeraeus capillatus* (LeConte) (Coleóptera: Curculionidae) en el Departamento de Córdoba-Colombia. Tem. Agric. 16: 23-35.
- Prena, J. 2009. A review of the species of *Geraeus pascoe* and *Linogeraeus casey* found in the continental United States (Coleoptera: Curculionidae: Baridinae). Coleopt. Bull. 63: 123-172.
- Reyes, J. L., M. M. Correa, V. Zamora, V. Ávila, M. F. Sánchez, and A. N. Rivera. 2021. Potenciales curculiónidos plaga en *Agave duranguensis* en el estado de Durango, México, pp. 44-48. In J. C. Herrera [ed.], Biodiversidad y Ecología Mexicana. Nuevos Conocimientos y Tecnologías para los Retos Actuales. Universidad Juárez del Estado de Durango, México.
- Rosas, M. V., J. J. Morrone, M. G. Del Rio, and A. A. Lanteri. 2011. Phylogenetic analysis of the *Pantomorus-Naupactus* complex (Coleoptera: Curculionidae: Entiminae) from North and Central America. Zootaxa 2780: 1-19.
- Salas, M. D., and A. Boradonenko. 2006. Insectos asociados al amaranto *Amaranthus hypocondriacus* L. (Amaranthaceae) en Irapuato, Guanajuato, México. Acta Univ. 16: 50-55.
- Sleeper, E. L. 1957. Notes on the genus *Macrorhoptus* Leconte (Coleoptera: Curculionidae, Anthonominae): 16. A contribution to the knowledge of the Curculionoidea. Ohio J. Sci. 57: 70-74.
- Soto, H. M. 2017a. Curculiónidos de México: Curculioninae y Baridinae, pp. 63-102. Introducción al Estudio de los Curculiónidos de México. Editorial Académica Española, España. ISBN: 978-3-659-65791-7
- Soto, M. 2017b. Nuevos registros de Attelabidae y Curculionidae para México (Coleoptera: Curculionoidea). Entomol. Mex. 4: 683-687.
- Sumano, D., V. H. Arias, and E. Capetillo. 2014. Primer registro de *Naupactus cervinus* (Coleoptera: Curculionidae) asociado al follaje de *Citrus latifolia* en el estado de Tabasco, México. Fitosen. 18: 49-50.

***Automeris moloneyi* Druce¹ Larvae Feed on Oil Palm Leaflets**

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Abstract. Oil palm, *Elaeis guineensis* Jacq., is one of the most important oil crops in Mexico. The study aimed to record herbivory, damage, and taxonomic identity of *Automeris moloneyi* Druce 1897 (Lepidoptera: Saturniidae) in nine commercial oil palm plantations at Tabasco, Mexico. In each plantation, 30 palm trees were selected in which *A. moloneyi* was searched for and collected every 3 months, from September 2019 to November 2020. At each sampling, the leaf surface of four fronds in the lower-middle part of the plant canopy was examined. *A. moloneyi* larvae fed on oil palm leaflets, abundance was low (0.003 to 0.103 larva per frond), and damage was localized (few plants). Most larvae per frond were recorded during the wettest time of the year in the study area. This is the first record of *A. moloneyi* in oil palm. The finding extends the geographical distribution of the species in the Neotropical region of the world. Probable factors that might be involved in low occurrence and damage of the species in the oil palm agroecosystem of the study area were discussed.

Introduction

The oil palm, *Elaeis guineensis* Jacq. (Arecaceae), is one of the most important oil crops in the world. In Mexico, the area cultivated in the last 10 years increased from 36,189 to 108,690 ha (SIAP 2019). The Mexican oil palm agribusiness has about 8,000 producers and generates 53,579 direct and indirect jobs (FEMEXPALMA 2020). The value of production in 2019 was 306.9 million pesos in the State of Tabasco and 1.685 billion pesos in Mexico (SIAP 2019).

Oil palm is attacked by several species of defoliator insects (Genty et al. 1978, Zenner de Polanía and Posada 1992, Aldana de la Torre 2010). Among them, four species of the genus *Automeris* damage oil palm in the Neotropical region of the world (Genty et al. 1978, Zenner de Polanía and Posada 1992, Couturier and Kahn 1993, Almeida de Oliveira et al. 2018, Barbosa da Silva 2019). Plantations in Brazil, Colombia, Ecuador, Peru, and Venezuela have been attacked (Genty et al. 1978).

¹Lepidoptera: Saturniidae: Hemileucinae

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The genus *Automeris* Hübner 1819 contains species of beautiful moths ranging in size from small to very large (Michener 1952). Their main characteristic is a large eyespot on the dorsal side of the hindwings and a rounded discal spot on the ventral view of the forewings (Lemaire 2002, Decaëns et al. 2021). The genus has 313 species worldwide (Kitching et al. 2018), of which 36 species have been recorded in Mexico (Balcázar-Lara and Beutelspacher-Baigts 2000).

Habitats of the genus *Automeris* are restricted to the Neotropical region of the world, from Canada to Argentina, except for six species that occur north of the Mexican border (Lemaire and Wolfe 1993, Lemaire 2002). *Automeris moloneyi* Druce 1897 (Lepidoptera: Saturniidae) has more limited geographical distribution. The species was first described from specimens collected in Belize (Druce 1891-1900), later reported in Guatemala (Monzon and Wolfe 1999), and more recently in Mexico (Lemaire 2002).

Little is known about herbivory and damage by *A. moloneyi*. In nature, it has only been recorded in sugarcane, *Saccharum officinarum* L. (Poaceae) (Lemaire 2002). During development of a study of phytophagous insects in oil palm, the objective was to document herbivory, damage, and taxonomic identity of *A. moloneyi* in nine commercial oil palm plantations at Tabasco, Mexico.

Materials and Methods

The study was done in commercial oil palm plantations at nine locations in the State of Tabasco, Mexico (Table 1). The climate of the State of Tabasco is warm-humid (Am or Af), with annual rainfall from 1,600 to 4,000 mm. Three dominant climatic seasons throughout the year are: 1) dry season (March to May) with warm

Table 1. Geographic Location and Age of Nine Oil Palm Plantations Sampled for Defoliator *Automeris moloneyi* in the State of Tabasco, Mexico.

Locality name	Geographical coordinates	Elevation (m above sea level)	Plantation age (years)*
El Encomadero, Huimanguillo	17.801631° N, 93.569280° W	34	5
José Ma. Pino Suárez, Huimanguillo	17.740312° N, 93.653986° W	47	3
Tecominoacán, Huimanguillo	17.926055° N, 93.574266° W	26	2
Aquíles Serdán, Macuspana	17.820179° N, 92.497559° W	7	5
Ceiba 1st sector, Tacotalpa	17.571983° N, 92.828683° W	25	6
Montaña, Jalapa	17.658796° N, 92.824866° W	16	4
Ignacio Allende 2nd sector, Teapa	17.551983° N, 92.992702° W	35	5
La Pita, Emiliano Zapata	17.520865° N, 91.590953° W	22	4
Emiliano Zapata 2nd sector, Tenosique	17.403630° N, 91.316436° W	57	4

*At the start of sampling

temperatures and low rainfall; 2) rainy season (June to September) defined by torrential downpours of short duration; and 3) "nortes" period (October to February) characterized by prevailing northerly winds and prolonged low-intensity rains (drizzle) associated with cloudy days (Larios-Romero and Hernández 1987).

At each locality, a 10-ha oil palm plot was selected in which *A. moloneyi* was searched for and collected every 3 months from September 2019 to November 2020. On each sampling date, 30 plants were randomly selected from a zigzag route along the harvesting paths (Calvache 1995, Pedigo et al. 2021). On each plant, the upper and lower sides of four fronds (facing north, south, east, and west) from the lower-middle part of the plant canopy (between fronds 17-25) were examined entirely. According to Calvache (1995), visual inspection of plants along harvesting paths was adequate to detect initial infestations by insect pests in oil palm. Defoliating larvae of Lepidoptera on palms prefer to feed on older fronds (Howard et al. 2001).

Larvae collected were put in plastic containers and taken to the Plant Health Laboratory of the Academic Division of Agricultural Sciences of the Universidad Juárez Autónoma de Tabasco. They were kept at a temperature of $28 \pm 2^\circ\text{C}$, $75 \pm 5\%$ relative humidity, and a photoperiod of 12 hours in an insect-rearing chamber in the laboratory. Larvae were fed oil palm leaflets until they pupated and then were preserved until adults emerged.

Emerged moths were killed by injecting 0.2 ml of 70% alcohol into the mesosternum of the thorax. They were then stored and dried in labeled paper envelopes for subsequent standard mounting that allowed optimal examination of the body and wings. The genitalia and eighth abdominal segment of males and females were prepared in a 10% potassium hydroxide solution to remove piliform scales and were preserved in 75% ethanol. Body morphology, wing ornamentation, and genitalia structure of males and females were compared with characteristics of the *Automeris randa* species-subgroup represented in Lemaire (2002).

The taxonomic identity of *A. moloneyi* adults was complemented with the description of the color pattern of the last larval instar and some observations of its habits and damage. The proportion of larvae per frond by locality and sampling period throughout the study was compared using a graph made using Microsoft Excel software (Henderson 2021).

Results and Discussion

Automeris moloneyi larvae feed on oil palm leaflets (Fig. 1A). Small larvae are gregarious on a single frond leaflet (Fig. 1B). As they grow, medium-sized larvae disperse to other leaflets on the frond, or even to other fronds on the plant. They are on upper fronds of the plant canopy of 2- to 3-year-old palms (Fig. 1B) and are on lower fronds (Figs. 1C,D) on 5- to 6-year-old palms. *A. moloneyi* were not abundant and damage was incipient in the study area, but attack by a few larvae can significantly reduce leaf area of a plant (Fig. 1A). Our finding is the first record of herbivory by the species on oil palm leaflets.

Little is known of the host plants and damage by *A. moloneyi*. It feeds on sugarcane, *Saccharum officinarum* L. (Poaceae), in the wild and has been reared on *Quercus* sp. (Fagaceae) in a laboratory (Lemaire 2002). In our study, field-collected larvae were successfully fed to the adult stage on leaflets of oil palm and coconut palm, *Cocos nucifera* L. (Arecaceae), as well as on leaves of Tó, *Calathea lutea* (Aubl.) E.Mey. ex Schult. (Marantaceae).

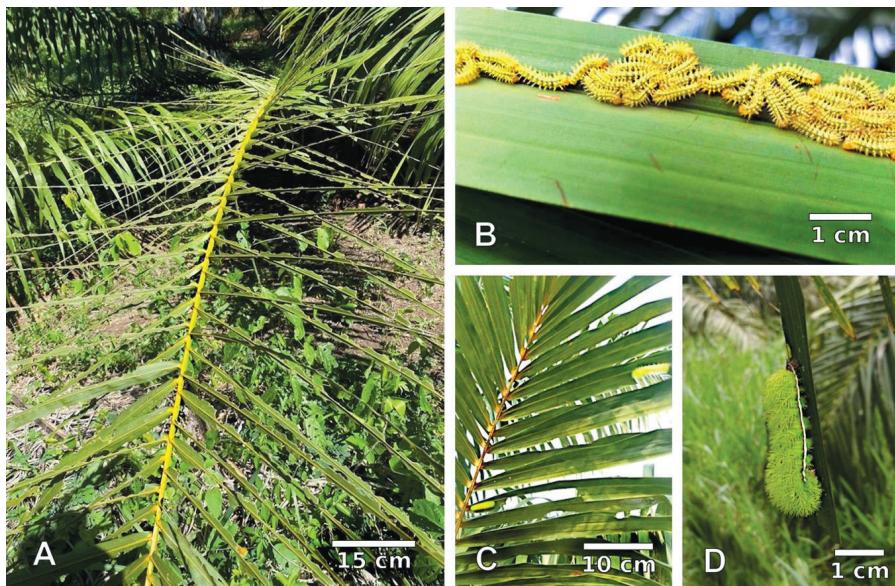


Fig. 1. Larvae of and damage by *Automeris moloneyi* on leaflets of oil palm (*Elaeis guineensis*). A) Larval damage to leaflets of a frond. B) Small larvae (gregarious) on a leaflet. C) Medium-sized larvae feeding on leaflets. D) Last-instar larvae on a leaflet.

Eggs of *A. moloneyi* are 1.7 x 1.4 x 0.8 mm and ovoid, smooth, and slightly flattened on both sides (Fig. 2A). They are deposited in groups of four to 35 eggs in a laboratory. At first, eggs are white but later turn yellowish and often are glued with an adhesive substance to the surface of a leaf blade. First-instar larvae are gregarious (Fig. 2B) and protected by scoli that have a mechanical defense function and stinging substances.

Last-instar larva can be 50 mm long (Fig. 2C). The head and rest of the body are green, covered dorsally and laterally by prominent urticating scoli. It has light orange spiracles. On the lateral part of the abdomen is a white subspiracular band from segment A1 to A10, bordered at the top and bottom with black. A black ventral spot between the prolegs of abdominal segments A1 to A7 is dotted with white (Fig. 2C). Prolegs have a black cross band and biordinal crochets arranged in cross bands. Before pupating, the larva builds a silken cocoon or puparium from dried fragments of palm leaflet (Fig. 2D).

The color of the thorax, wings, and abdomen of an adult is similar to that described by Lemaire (2002). Females are larger and more robust than males (wingspan 80-86 mm and 55-74 mm, respectively) (Fig. 3). Antennae of a female are filiform, while those of males are plumose (quadripectinate). Mouthparts of both sexes are degenerate, and the head is small (Lampe 2010).

Forewings of females and males are not elongated but have a rounded apex and straight or slightly convex outer margin. Forewings of females are dark brown, while those of males are light brown. A narrow dark-brown transverse line from the

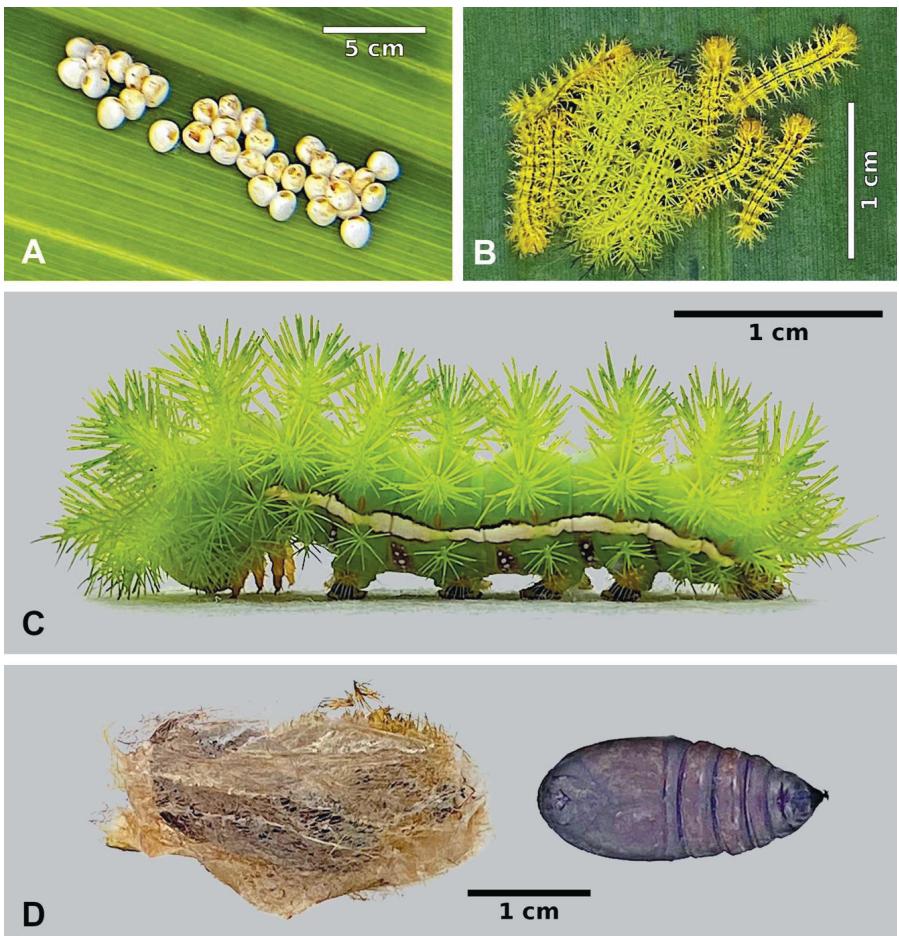


Fig. 2. Developmental stages of *A. moloneyi*. A) Eggs, B) gregarious larvae, C) last-instar larva, and D) puparium and pupa.

tornus (dorsal margin) to the apex (costal margin) is in the dorsal view of the forewings of both sexes. At the base of veins M1 to M3 is an irregular dark-brown marking, which in the ventral view corresponds to a black discal spot with a white dot in the center. A dorsal view of the hindwings shows a large, slightly oval eyespot composed of a black pupil (crossed by a line of white scales) and a brown iris surrounded by a concentric black ring which in turn is bordered by a ring of yellow scales that enhance its size and visibility. A white dot is visible in the ventral view of the eyespot. The submarginal area of the hindwings (between the tornus and apex) features a wavy black line bordered by yellow scales. The marginal area of the hindwings has a grayish-pink band that contrasts with the color of the rest of the wings.

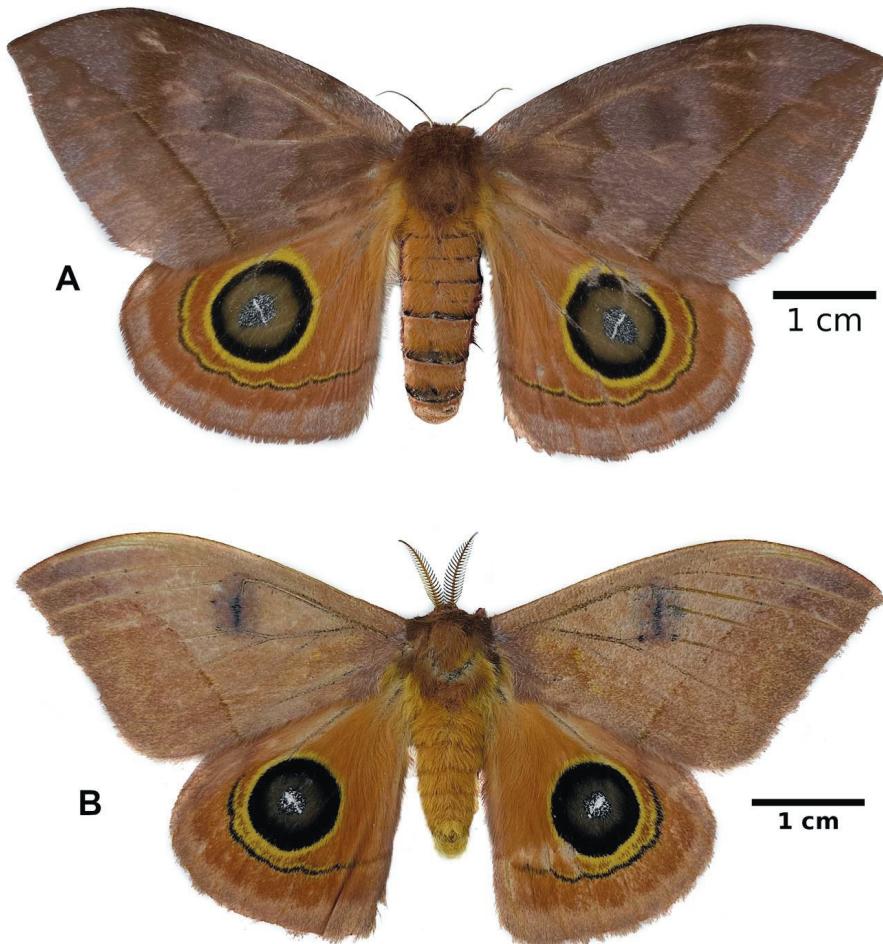


Fig. 3. Adults of *Automeris moloneyi*. A) Female and B) male.

In a comparative study of genitalia of males and females (Fig. 4), not a single morphological trait different from that of *A. moloneyi* species described by Lemaire (2002) was found. *Automeris moloneyi* has limited geographical distribution in the world. It was found in Belize (Druce 1891-1900), Guatemala (Monzon and Wolfe 1999), and Mexico (Lemaire 2002). In Mexico, it was recorded at Cozumel, Lázaro Cárdenas, and Puerto Morelos, Quintana Roo (DGRU-UNAM 2022), and at Calakmul, Campeche, and Salto de Agua, Chiapas (Ratnasingham and Hebert 2007). Our finding of the species at Tabasco extends the geographical distribution of *A. moloneyi* in the Neotropical region of the world.

In total, 134 larvae of *A. moloneyi* were found: 62 larvae were at the oil palm farm of José Ma. Pino Suárez ejido (0.103 larva/frond), 31 larvae at El Encuentro ejido (0.052 larva/frond), 13 larvae at the Emiliano Zapata 2nd sector ejido (0.022

larva/frond), 11 larvae at the Ceiba 1st sector ejido (0.018 larva/frond), and 17 larvae at the five other localities (Fig. 5). Thus, most larvae were collected at José Ma. Pino Suárez and El Encomedero from June–November, which is the wettest time of the year in the State of Tabasco (Larios-Romero and Hernández 1987).

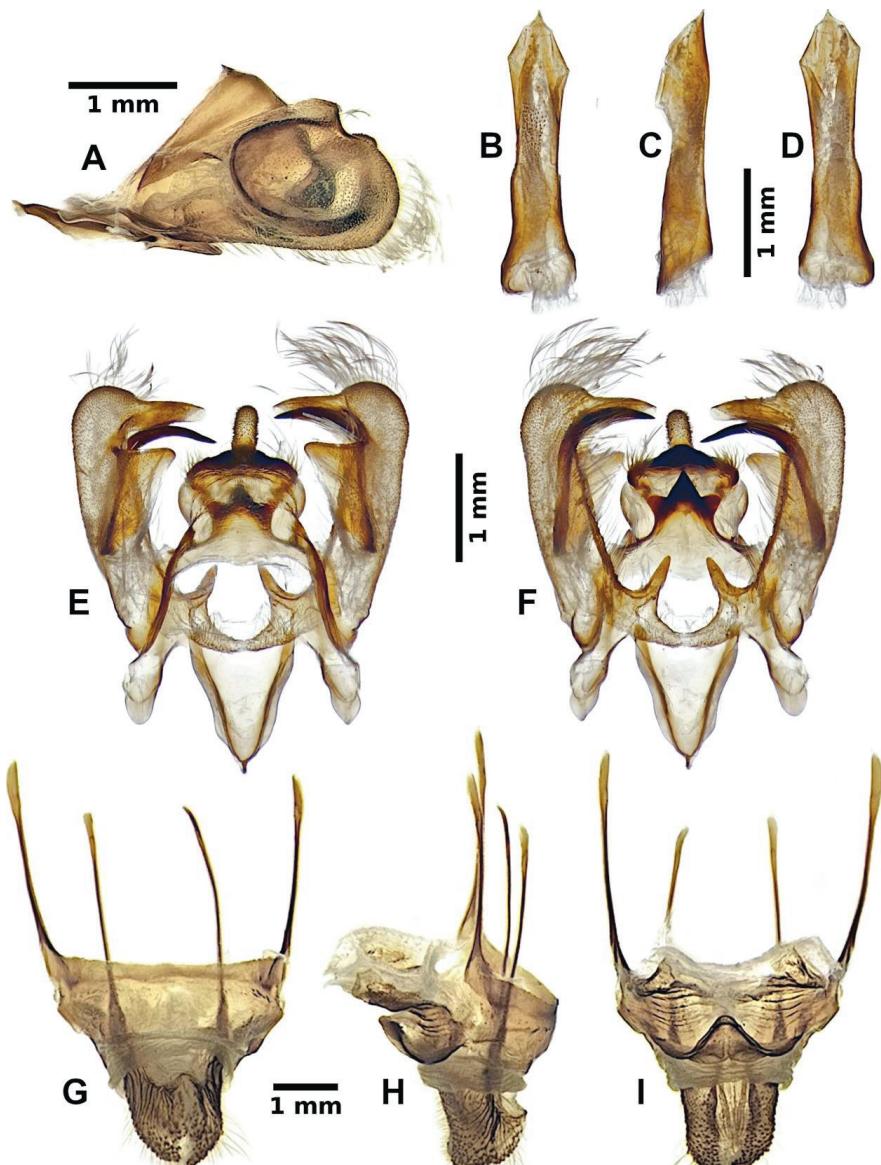


Fig. 4. Genitalia of *Automeris moloneyi*. A) Left lateral view of male genitalia. B-D) Dorsal, lateral, and ventral views of the aedeagus. E, F) Dorsal and ventral views of male genitalia. G-I) Dorsal, lateral, and ventral views of female genitalia.

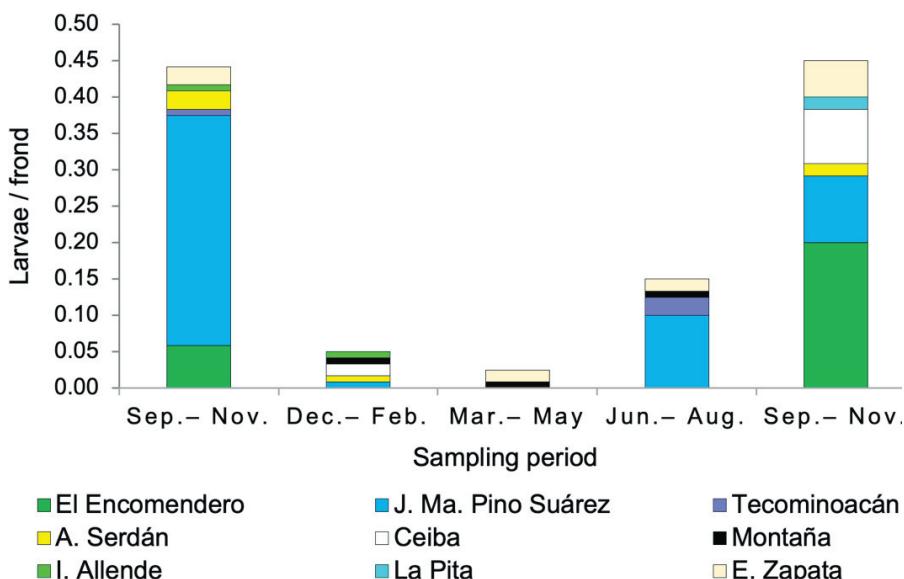


Fig. 5. Cumulative rates of *Automeris moloneyi* larvae by sampling period from September 2019 through November 2020 at commercial oil palm plantations at nine locations in the State of Tabasco.

Of the specimens, 40.3% were gregarious second- or third-instar larvae. A first group of 38 second-instar larvae was at the J. María Pino Suárez ejido, on 12 October 2019; a second group of 16 third-instar larvae was at El Encomendero ejido on 22 September 2020. Finding small larvae at that time of year might be related to reproductive activity of *A. moloneyi* in oil palm plantations at Huimanguillo, Tabasco.

According to bibliographical information, four species of *Automeris* (*A. liberia* Cramer, *A. cinctistriga* Felder & Rogenhofer, *A. vividior* Draudt, and *A. bilineata* Walker) defoliate oil palm plantations in the Neotropical region of the world (Genty et al. 1978, Zenner de Polanía and Posada 1992, Couturier and Kahn 1993, Almeida de Oliveira et al. 2018, Barbosa da Silva 2019). The most important species is *A. liberia* that attacks *E. guineensis* in Brazil, Colombia, Ecuador, Peru, and Venezuela (Genty et al. 1978). A single larva of *A. liberia* can destroy 1,000 cm² of frond (four leaflets) and is especially damaging in crops that are 1 to 3 years old.

In addition to oil palm, *A. liberia* and *A. cinctistriga* attack other palm species in some parts of tropical Americas (Howard et al. 2001). For example, *A. cinctistriga* attacks coconut palm in Brazil (Rollemburg et al. 2002). Coconut palm plants and other species of Arecaceae are in the State of Tabasco, Mexico (Magaña and González 2017), in some cases surrounding oil palm plantations and, in effect, could be host plants of *A. moloneyi*.

Throughout the study period, *A. moloneyi* was attacked by three species of natural enemies -- a predatory bug of Pentatomidae, a wasp of Ichneumonidae, and a fly of Tachinidae (data to be published). Predation and parasitism by the species exert important and effective natural biological control that manages abundance of *A. moloneyi* in the study area.

In conclusion, *A. moloneyi* feeds on oil palm leaflets in plantations at Tabasco, Mexico. This is the first finding of herbivory by larvae of the species, a record that expands its geographical distribution in the Neotropical region of the world. Abundance and damage found were incipient and localized (few plants), probably because of entomophagous insects that exert good natural control. It is necessary to expand the study of herbivory by *A. moloneyi* to other plant species in the oil palm agroecosystem of the study area, as well as identify entomophagous insects and their parasitism or predation rates. Knowledge of interrelationships of the defoliator species with its host plants and natural enemies will be useful for defining management practices to control outbreaks in the oil palm agroecosystem.

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References Cited

- Aldana de la Torre, R. C., J. A. Aldana de la Torre., H. Calvache G., and P. N. Franco B. 2010. Manual de Plagas de la Palma de Aceite en Colombia. 4a edición. CENIPALMA, Bogotá, Colombia.
- Almeida de Oliveira, T., W. P. Lemos, R. S. Tinoco, J. C. Barbosa, and I. C. F. Martins. 2018. Validation of the sampling methodology for *Opsiphanes invirae* caterpillars on oil palm plantations in the Brazilian Amazon. Crop Protection Ciéncia Rural. <https://doi.org/10.1590/0103-8478cr20170742>.
- Balcázar-Lara, M. A., and C. R. Beutelspacher-Baigts. 2000. Saturniidae (Lepidoptera), pp. 501-513. In J. B. Llorente, E. S. González y N. Papavero [eds.], Biodiversidad, Taxonomía y Biogeografía de Artrópodos de México: Hacia una Síntesis de su Conocimiento, vol. II. Universidad Nacional Autónoma de México, México, D. F.
- Barbosa da Silva, K., C. Barbosa da Silva, K. A. L. Ribeiro, J. McLaine, J. Duarte de Freitas, G. Sanchez Chia, R. S. Tinôco, J. Gomes da Costa, H. F. Goulart, and A. E. Goulart. 2019. Morphology and distribution of antennal sensilla of *Automeris libera* T (Lepidoptera: Saturniidae). Micron 123 <https://doi.org/10.1016/j.micron.2019.102682>
- Calvache, H. G. 1995. Manejo integrado de plagas de la palma de aceite. Palmas 16: 255-264.
- Couturier, G., and F. Kahn. 1993. A new pest of the African oil palm in the Neotropics: *Periphoba hircia* (Lepidoptera Saturniidae Hemileucinae). Principes 37: 228-229.
- Decaëns, T., F. Bénéluz, L. Ballesteros-Mejia, D. Bonilla, and R. Rougerie. 2021. Description of three new species of *Automeris* Hübner, 1819 from Colombia and Brazil (Lepidoptera, Saturniidae, Hemileucinae). ZooKeys 1031: 183-204.
- DGRU-UNAM (Dirección General de Repositorios Universitarios - Universidad Nacional Autónoma de México). 2022. Portal de Datos Abiertos UNAM, Colecciones Biológicas: *Automeris moloneyi* Druce, 1897. <https://datosabiertos.unam.mx/IBUNAM:CNIN:LE1522> Accessed 19 April 2022.

- Druce, H. 1891-1900. Biología Centrali-Americana: Lepidoptera-Heterocera. Vol. II. Published for the editors by R. H. Porter. London.
- FEMEXPALMA (Federación Mexicana de Palma de Aceite). 2020. Anuario Estadístico 2020. Villahermosa, Tabasco, Mexico.
- Genty, P., Desmier De Chenon Roch, and Morin Jean-Paul. 1978. Les ravageurs du palmier à huile en Amérique Latine. Oléagineux 33: 325-419.
- Henderson, P. A. 2021. Southwood's Ecological Methods. 5th ed. Oxford University Press, UK.
- Howard, F. W., D. Moore, R. M. Giblin-Davis, and R. G. Abad. 2001. Insects on Palms. CAB International, Wallingford, UK.
- Kitching, I. J., R. Rougerie, A. Zwick, C. A. Hamilton, R. A. St Laurent, S. Naumann, L. Ballesteros M., and A. Y. Kawahara. 2018. A global checklist of the Bombycoidea (Insecta: Lepidoptera). Biodiversity Data Journal 6: e22236. <https://doi.org/10.3897/BDJ.6.e22236> Accessed 12 October 2021.
- Lampe, R. E. J. 2010. Saturniidae of the World: Their Life Stages from Eggs to the Adults. Verlag Dr. Friedrich Pfeil, München, Germany.
- Larios-Romero, J., and J. Hernández. 1987. Condiciones ambientales para la agricultura en el estado de Tabasco. Revista de Geografía Agrícola 13-14: 33-42.
- Lemaire, C. 2002. The Saturniidae of America. Hemileucinae. Antiquariat Geock & Evers. Keltern, Germany.
- Lemaire, C., and K. L. Wolfe. 1993. Two new *Automeris* from Western Mexico (Lepidoptera: Saturniidae: Hemileucinae). Tropical Lepidoptera 4: 39-44.
- Magaña A., M. A., and A. González H. 2017. Estudio taxonómico de la familia Arecacea en el municipio de Macuspana, Tabasco, México. Kuxulkab' 23: 5-15.
- Michener, C. D. 1952. The Saturniidae (Lepidoptera) of the Western Hemisphere: morphology, phylogeny, and classification. Bull. Am. Mus. Nat. Hist. 98: 335-502.
- Monzon S., J., and K. L. Wolfe. (1999). Lista preliminar de las especies de la familia del "gusano de pino" en Guatemala (Lepidoptera: Saturniidae). Revista Nicaraguense de Entomología 48: 29-35.
- Pedigo, L. P., M. E. Rice, and R. K. Krell. 2021. Entomology and Pest Management. 7th ed. Waveland Press, Long Grove, IL.
- Ratnasingham, S., and P. D. N. Hebert. 2007. BOLD: The Barcode of Life Data System (<https://www.boldsystems.org/index.php>). Molecular Ecology Notes 7: 355-364.
- Rollemburg, F. H., J. M. S. Ferreira, and L. A. Siqueira. 2002. Sistema de produção para a Cultura do Coqueiro. Empresa Brasileira de Pesquisa Agropecuária (EMBRAPA). Aracajú, Sergipe, Brasil.
- SIAP (Servicio de Información Agroalimentaria y Pesquera). 2019. Anuario estadístico de la producción agrícola. <https://nube.siap.gob.mx/cierreagricola/> Accessed 12 March 2022.
- Zenner de Polanía, I., and F. J. Posada F. 1992. Manejo de Insectos, Plagas y Benéficos de la Palma Africana. Produmedios. Bogotá, Colombia.

Wasp Moths (Erebidae) of the Lepidoptera Collection at the University of Colima, Mexico

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Abstract. Ctenuchina and Euchromiina moths (Erebidae: Arctiinae) in the entomological collection at the University of Colima were revised and identified to species. A total of 603 specimens belonging to 40 species were examined. All wasp-moth species belonged to two subtribes: Ctenuchina (three species) and Euchromiina (37 species). A species list with collecting details is presented.

Resumen. Las polillas Ctenuchina y Euchromiina (Erebidae: Arctiinae) depositadas en la colección entomológica de la Universidad de Colima fueron revisadas e identificadas. Un total de 603 ejemplares pertenecientes a 40 especies fueron examinados. Estos pertenecen a dos subtribus: Ctenuchina (tres especies) y Euchromiina (37 especies). Una lista de especies con detalles de recolecta se presenta en este trabajo.

Introduction

The Biological and Agricultural Sciences School (FCBA, after its Spanish name) of Colima University (Universidad de Colima, UCOL) was founded in 1976 as the Superior School of Agriculture. Thirty years later, in 2004, the Lepidoptera Collection was created at the Tecomán campus ($N18^{\circ}54'33'' W103^{\circ}53'37''$) of UCOL. One of the main objectives of the newly founded insect collection was surveying and describing lepidopterans, especially moths in the diverse ecosystems of the State of Colima, Mexico, as well as adjacent regions in western Mexico. Currently, 35,000 specimens are in the Lepidoptera collection. Despite its small size, the UCOL Lepidoptera collection is the only important institutional insect collection along the Mexican Pacific Coast.

In Mexico, 14,385 species of Lepidoptera in 32 families have been recorded (Heppner 2002). Species richness of the subtribes Ctenuchina and Euchromiina (Noctuoidea: Erebidae: Arctiinae: Arctiini) was estimated at 2,532 species, with 2,475 being exclusively Neotropical (Heppner 1991) while only 36 are Nearctic (Heppner 1991, Lafontaine and Fibiger 2006, Lafontaine and Schmidt 2010). Simmons et al. (2012) suggested as many as 3,000 species. So far, 240 species are known from Mexico (Hernández-Baz 2012, 2013, 2016), 128 in 39 genera of Ctenuchina, while

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the remaining 112 species are in the 31 genera of Euchromiina, with 10 endemics (Hernández-Baz 1992, 2008, 2009, 2010, 2011a,b, 2013). Most species fly mainly during the day (Hernández-Baz et al. 2012, 2013), although some have been captured at lights (Hernández-Baz and Bailey 2006).

Besides lacking or having incomplete faunistic inventories of moths, knowledge of their geographical distribution is absent, especially for Ctenuchina and Euchromiina. This is especially true in Mexico, where surveys for most Lepidoptera families are uncommon. However, during the last 30 years one of us (FHB) has been attempting to complete a national survey of all Ctenuchina and Euchromiina (Lepidoptera: Erebidae), gathering data and information for all known and collected species from Mexico in the last 200 years (Hernández-Baz 2012). Since Mexican wasp moths are poorly studied, especially from the Pacific slope, our aim is to provide a list of species in the moth collection of the Facultad de Biología de la Universidad de Colima, México.

Materials and Methods

Information provided is from Ctenuchina and Euchromiina specimens at the Lepidoptera collection (UCOL) at Facultad de Ciencias Biológicas of Colima University, México. All registries obtained are in a database prepared in Biota V 2.0 (Coldwell 2000). Species were identified by using Hampson (1898, 1914), Draudt (1917), Dietz and Duckworth (1976), Dietz (1994), and Hernández-Baz (1992, 2012).

All locality registries were geo-referenced based on the names catalog at the National Institute of Geography and Informatics of México and a 1:250,000-scale topographic map of the country (INEGI 2012). All localities were confirmed with <http://www.google.com/earth>. The main collecting localities of wasp moths at Colima were designed with Arc View 2.0 (ESRI 1998)

Results and Discussion

In total, 603 specimens of 40 species collected at several localities of the State of Colima and deposited at UCOL were studied (Table 1). Among them were 38 specimens of three species in three genera and 38 specimens of Ctenuchina (Fig. 1); the other 37 taxa were Euchromiina with 21 genera and 565 specimens (Figs. 1, 2). Hernández-Baz (2012, 2016) previously registered 28 species for the State of Colima. Herein, the number increased 30%. Twenty-three collecting sites were listed, including 13 at Colima, ranging from 500 to 2,400 m above sea level. They belong to different ecological regions ranging from commercial palm groves, lowland oak woodlands, and mountain pine forests. Collecting sites at Colima cover 50% of its territory. Seven sites were in the State of Sonora, and one site were in each of Jalisco, Querétaro, and San Luis Potosí states (Table 2).

A well-represented annual distribution of Ctenuchina and Euchromiina species was found in the curatorial data in the UCOL collection. Richness and abundance peaked between August and October, at the end of summer and beginning of fall at Colima. Few species were found during winter (December, January, and February) (Fig. 3). A similar situation occurred in the central region of the State of Veracruz at approximately the same latitude as Colima (Hernández-Baz 1993).

Only two species *Cosmosoma theutras cingulatum* Butler with 66 specimens and *Macrocneme chrysitis* (Guérin-Menéville) with 59 were at Colima throughout the year. Both species are multivoltine and more abundant at the end of the summer and

Table 1. Ctenuchina and Euchromiina Moths (Lepidoptera: Erebidae) in the Scientific Collection of Lepidoptera, University de Colima (UCOL), Mexico

N u m b	Month of 2001-2009 years											
	J	F	M	A	M	J	J	A	S	O	N	D
Ctenuchina:												
5 <i>Ctenucha venosa</i> Walker								7	4	3	21	35
19 <i>Philaros rubriceps</i> (Walker)								1				1
20 <i>Theages flavigaput</i> (Hampson)									1	1		2
Specimens of Ctenuchina								1	7	4	4	22
Euchromiina:												38
1 <i>Abrochia leovazquezae</i> (Pérez y Sánchez)								1				1
2 <i>Aclyta heber</i> (Cramer)									3	2	5	1
21 <i>Apeplopoda ochracea</i> (Felder)						3	1	4		4	1	13
22 <i>Chrysocale principalis</i> (Walker)									7			7
3 <i>Correbia undulata</i> (Druce)									1		5	6
4 <i>Correbida germana</i> (Rothschild)								1	1	2	1	5
23 <i>Cosmosoma auge</i> (Linnaeus)			1		2	13	9	1	5	1		32
24 <i>Cosmosoma festiva</i> (Walker)									1			1
25 <i>Cosmosoma teuthras cingulatum</i> Butler	2	7	9	1	5	7	1	5	10	12	6	1
26 <i>Dycladia correbooides</i> Felder					8	1	6	1	2	5	6	29
6 <i>Eucereon erythrolepis</i> Dyar						1			5	3	3	12
7 <i>Eucereon latifascia</i> (Walker)							1	1	1	1	1	5
8 <i>Eucereon leria</i> Druce							1			2	4	8
9 <i>Eucereon maia</i> Druce									1			1
10 <i>Eucereon myrina</i> Druce	1	1	4			4	2	15	8	4	1	40
11 <i>Eucereon phaeoproctum</i> (Hampson)					1	1	2	2	2	1	2	1
12 <i>Eucereon punctata</i> (Guérin-Ménéville)									2	3		5
13 <i>Eucereon tarona</i> Hampson	1						1	1	1			3
14 <i>Eucereon tripunctatum</i> Druce	1	1					1	2	4	3	3	1
15 <i>Eucereon xanthoperas</i> Hampson								1	1	3	1	6
16 <i>Horama panthalon texana</i> (Grote)						1		16	8	11	2	38
27 <i>Isanthrene perboscii</i> (Guérin-Menéville)					1	3	3		2	6	12	3
28 <i>Leucotmemis nexa</i> (Herrick- Schäffer)					1	1			1			3
29 <i>Leucotmemis pardalimacula</i> Dyar										1		1
30 <i>Loxophlebia imitata</i> (Druce)					1	1	1	1	1	2		7
31 <i>Macrocneme chrysitis</i> (Guérin-Meneville)	2	1	3	1	1	1	7	10	7	8	15	3
17 <i>Nelpha carolina</i> H. Edwards					1					1	5	1
18 <i>Nelpha rogersi</i> (Druce)					1		2	1	6	3	3	1
32 <i>Poliopastea clavipes</i> (Boisduval)									1	1	1	3
33 <i>Poliopastea laciades</i> (Schaus)									1			1
34 <i>Psilopleura vittata</i> (Walker)							16	17	18	8	1	1
35 <i>Rhynchopyga flavicollis</i> (Druce)							1					1
36 <i>Scena propylea</i> Druce					2	1					1	4
37 <i>Sphecosoma felderii</i> (Druce)										1		1
38 <i>Syntomeida hampsonii</i> Barnes								2	3	3		8
39 <i>Syntomeida melanthus albifasciata</i> Butler					10	10	2	2	1	2	4	8
40 <i>Syntomeida syntomoides</i> (Boisduval)										1		1
Specimens of Euchromiina	9	10	31	7	30	55	62	101	81	83	74	22
Total number of specimens	9	10	31	7	31	55	69	105	85	105	74	22
											603	

during fall. Nine of 40 registered species collected only once during 2001-2009 were Ctenuchina: *Philaros rubriceps* (Walker), and Euchromiina: *Abrochia leovazquezae* (Pérez and Sánchez), *Cosmosoma festiva* (Walker), *Eucereon maia* Druce, *Leucotmemis pardalimacula* Dyar, *Poliopastea laciades* (Schaus), *Rhynchopyga flavicollis* (Druce), *Sphecosoma felderii* (Druce), and *Syntomeida syntomoides* (Boisduval). All were collected from May to October, months with high heat and rainfall.

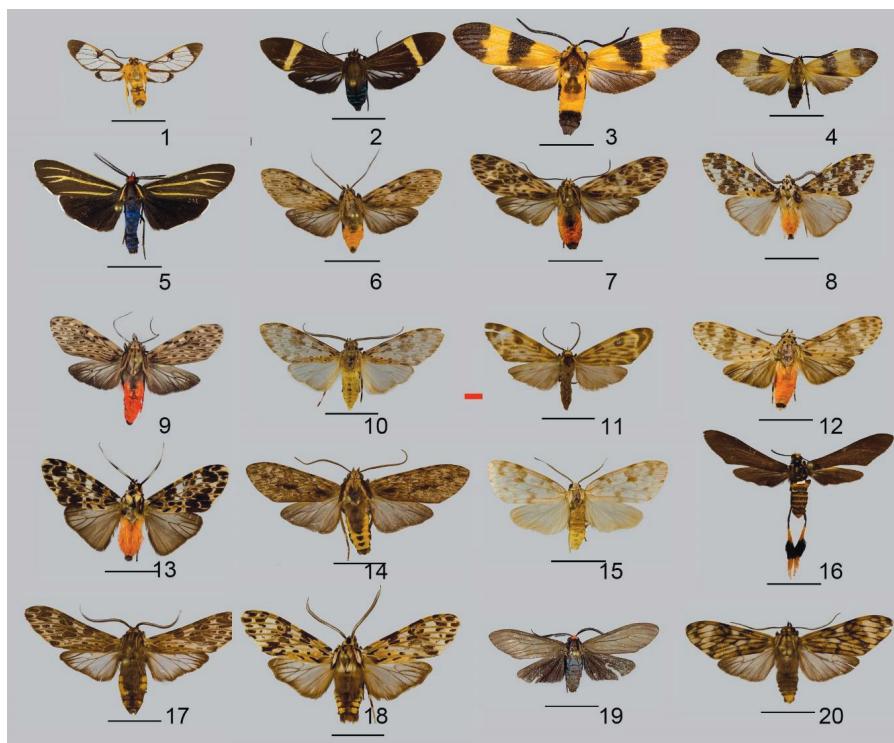


Fig. 1. Images of wasp moths (Ctenuchina and Euchromiina) deposited at the Lepidoptera collection, Universidad de Colima, Mexico, and collected during 2001-2009. 1. *Abrochia leovazquezae* (Pérez y Sánchez), 2. *Aclytia heber* (Cramer), 3. *Correbia undulata* (Druce), 4. *Correbidia germana* (Rothschild), 5. *Ctenucha venosa* Walker (Ctenuchina), 6. *Eucereon erythrolepis* Dyar, 7. *Eucereon latifascia* (Walker), 8. *Eucereon leria* Druce, 9. *Eucereon maia* Druce, 10. *Eucereon myrina* Druce, 11. *Eucereon phaeopuctum* (Hampson), 12. *Eucereon punctata* (Guérin-Ménville), 13. *Eucereon tarona* Hampson, 14. *Eucereon tripunctatum* Druce, 15. *Eucereon xanthopera* Hampson, 16. *Horama panthalon texana* (Grote), 17. *Nelpha carolina* H. Edwards, 18. *Nelpha rogersi* (Druce), 19. *Philaros rubriceps* (Walker), (Ctenuchina), 20. *Theages flavicaput* (Hampson) (Ctenuchina). Photos by Fernando Hernández-Baz and Manuel Balcázar Lara.

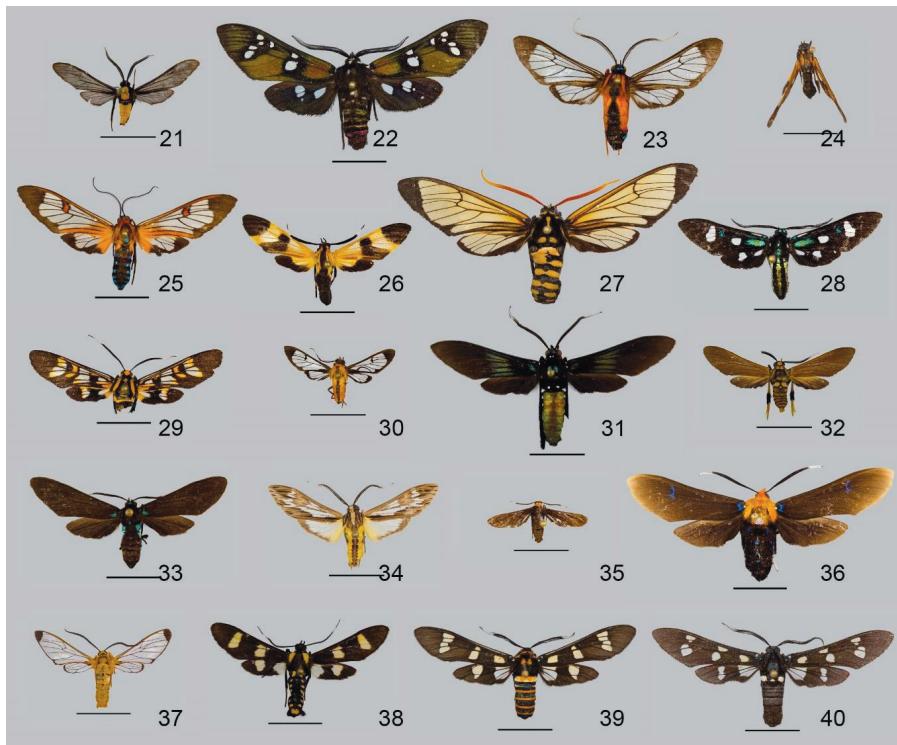


Fig. 2. Images of wasp moths (Euchromina) deposited at the Lepidoptera collection, Universidad de Colima, Mexico, and collected during 2001-2009. 21. *Apeplopoda ochracea* (Felder), 22. *Chrysocale principalis* (Walker), 23. *Cosmosoma auge* (Linnaeus), 24. *Cosmosoma festiva* (Walker), 25. *Cosmosoma teuthras cingulatum* Butler, 26. *Dycladia correbooides* Felder, 27. *Isanthrene perbosci* (Guérin-Menéville), 28. *Leucotmemis nexa* (Herrick-Schäffer), 29. *Leucotmemis pardalimacula* Dyar, 30. *Loxophlebia imitata* (Druce), 31. *Macrocneme chrysitis* (Guérin-Meneville), 32. *Polliopastea clavipes* (Boisduval), 33. *Polliopastea laclades* (Schaus), 34. *Psilopleura vittata* (Walker), 35. *Rhynchopyga flavicollis* (Druce), 36. *Scena propylea* Druce, 37. *Sphecosoma felderi* (Druce), 38. *Syntomeida hampsonii* Barnes, 39. *Syntomeida melanthus albifasciata* Butler, 40. *Syntomeida syntomoides* (Boisduval). Photos by Fernando Hernández-Baz and Manuel Balcázar Lara.

Table 2. Collecting Sites for Ctenuchina and Euchromiina Moths (Erebidae) at the State of Colima, México, Based on Data at Entomological Collection, Universidad de Colima (UCOL)

State	County	Locality and altitude	Latitude	Longitude
Colima	Colima	Colima, 500 m	19° 14' 35"	103° 43' 49"
Colima	Comala	La María, 1250 m	19° 27' 28"	103° 42' 21"
Colima	Comala	Zaculpan, 683 m	19° 21' 30"	103° 50' 15"
Colima	Comala	Ecoparque Nogueras, 656 m	19° 19' 24"	103° 44' 24"

Colima	Comala	Mesa de la Yerbabuena, 1660 m	19° 28' 47"	103° 40' 56"
Colima	Comala	Laguna La María, 1250 m	19° 27' 46"	103° 42' 29"
Colima	Coquimatlan	500 m E de el Colmo, 492 m	19° 09' 10"	104° 00' 02"
Colima	Ixtlahuacan	Gruta San Gabriel, 503 m	18° 54' 25"	103° 44' 24"
Colima	Minatitlán	Ocotillo, 2180 m	19° 26' 40"	103° 57' 18"
Colima	Minatitlán	500 m S de Agua fría, 742 m	19° 18' 07"	104° 03' 58"
Colima	Minatitlán	El Terrero, 2 km al NE., 2400 m	19° 27' 12"	103° 56' 34"
Colima	Tecomán	A 300 NE de la Salada, 355 m	19° 03' 24"	103° 47' 03"
Colima	Villa de Álvarez	1.5 km al SW de Zaculapan, 683 m	19° 14' 35"	103° 43' 49"
Jalisco	Pihuamo	La Escondida, 351 m	19° 02' 30"	103° 28' 03"
Querétaro	Querétaro	El lobo km 237 Carr. 120 Xilitla a Jalpan al Oeste de San Luis Potosí, 1626 m	21° 15' 46"	99° 10' 48"
San Luis Potosí	Tamazuchale	Tamanzuchale, 140 m	21° 15' 15"	98° 47' 16"
Sonora	Yécora	La Palmita, 1486 m	28° 22' 18"	109° 03' 52"
Sonora	Yécora	T. micro ondas de Yécora, 2200 m	28° 22' 04"	109° 02' 03"
Sonora	Yécora	Río Yécora, 1545 m	28° 22' 27"	108° 55' 14"
Sonora	Yécora	Arroyo La Culebra, 1650 m	28° 22' 37"	108° 53' 45"
Sonora	Yécora	El Ranchito, 1516 m	28° 23' 33"	108° 54' 40"
Sonora	Yécora	5 km SW de Santa Rosa, 824 m	28° 26' 33"	109° 06' 38"
Sonora	Yécora	Mina Trinidad, ca, 1320 m	28° 28' 21"	109° 02' 31"

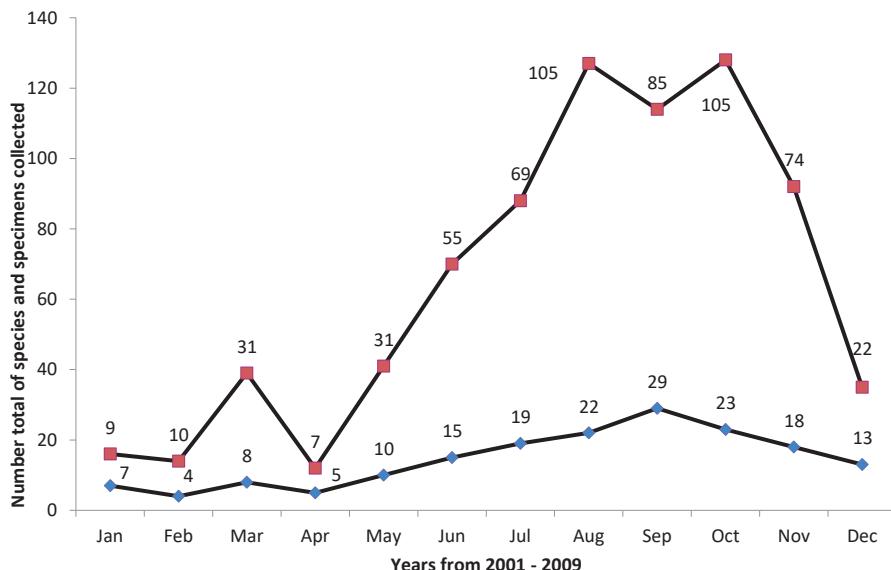


Fig 3. Total number of specimens (Ctenuchina and Euchromina) collected in the State of Colima, Mexico and deposited at the Lepidoptera collection, Universidad de Colima, Mexico. Curve with diamonds shows richness and curve with squares shows abundance of wasp moths collected during 2001-2009.

In general, wasp moths show a high level of synomization, followed by poor description of most taxa, making it a taxonomically confused group. There is a poor inventory record, limited knowledge of geographical distribution or description of life cycles. Besides collecting, we are studying wasp moths in scientific collections throughout Mexico, with the purpose of a) knowing all species found in Mexico, complementing all known information of each taxon found in curatorial data, and including all information obtained from "POLILLA" database; and b) generating an image bank of wasp moths to be put on WWW (World Web Wide) through the electronic page of the Faculty of Biology of Veracruz University in Mexico, providing access to anyone interested in this group of moths. The information provided is of the utmost relevance since the UCOL has one of the most complete collections of Lepidoptera inhabiting the Mexican Pacific Coast regions, including the Western section of the Western Volcanic Transversal Range.

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References Cited

- Colwell, R. K. 2000. EstimateS statistical estimation of species richness and shared species from sample. (Software and User's Guide). Version 7.52, Department of Ecology and Evolutionary Biology, University of Connecticut, <http://viceroy.eeb.uconn.edu/estimates/> Accessed 12 August 2021.
- Dietz IV, R. E. 1994. Systematics and Biology of the genus *Macrocneme* Hübner (Lepidoptera: Ctenuchinae). University of California Publications in Entomology 113: 1-121.
- Dietz IV, R. E., and W. R. Duckworth. 1976. A review of the genus *Horama* Hübner and reestablishment of the genus *Polioptastea* Hampson (Lepidoptera: Ctenuchinae), pp. 1-53. Smithsonian Contributions to Zoology No. 215. Smithsonian Institution Press, Washington, DC.
- Draudt, M. 1917. Syntomidae, pp. 33-230, In A. Seitz [ed.], Die Gross-Schmetterlinge der Erde. II. Abteilung: Die Gross-Schmetterlinge des Amerikanischen Faunengebietes, 6 Band, Die Amerikanischen Spinner und Schwärrmer. Alfred Kernen, Stuttgart.
- ESRI (Environmental Systems Research Institute, Inc.). 1998. Introduction to Arc View. GIS 3.2.
- Hampson, G. F. 1898. Catalogue of the Lepidoptera Phaleana in the Collection of the British Museum. British Museum Natural History, Longwans and Co., London. Vol. 1. XXII + 559 pp. 17 pls.
- Hampson, G. F. 1914. Catalogue of the Lepidoptera Phalaena in the Collection of the British Museum. British Museum Natural History, Longwans and Co., London. Suppl., Vol. I. XXVIII + 858 pp. + 41 plates.
- Heppner, J. B. 1991. Faunal regions and the diversity of Lepidoptera. Tropical Lepidoptera 2 (Suppl. 1): 1-85.
- Heppner, J. B. 2002. Mexican Lepidoptera biodiversity. Insecta Mundi 16: 171-190.
- Hernández-Baz, F. 1992. Catálogo de los Ctenuchidae (Insecta: Lepidoptera: Heterocera) de México. Bol. Soc. Mex. Lep. Nueva Serie 2: 19-47.

- Hernández-Baz, F. 1993. La fauna de mariposas (Lepidoptera: Rhopalocera) de Xalapa, Veracruz, México. *La Ciencia y El Hombre* 14: 55-87.
- Hernández-Baz, F. 2008. Mariposas de la familia Arctiidae de Aguascalientes, México, pp. 130-131. In H. Ávila, E. D. Melgarejo, y A. A. Cruz [eds.], *La Biodiversidad de Aguascalientes, Estudio de Estado*, Conabio, Instituto del Medio Ambiente del Estado de Aguascalientes, Universidad Autónoma de Aguascalientes, México.
- Hernández-Baz, F. 2009. Mariposas Arctiidae, pp. 109-112, 409-410. In G. Ceballos, R. List, G. Garduño, C. R. López, M. J. Muñozcano-Quintana, E. Collado, y J. E. San Román. [comp.], *La Diversidad Biológica del Estado de México. Estudio de Estado*. Conabio, Gobierno del Estado de México, México.
- Hernández-Baz, F. 2010. Arctiidae: Palomillas nocturnas, pp. 245-246. In R. Durán y M. Méndez [eds.], *Biodiversidad y Desarrollo Humano en Yucatán*, CICY, RPD-FMAM, CONABIO, SEDEMA, México.
- Hernández-Baz, F. 2011a. Palomillas nocturnas Arctiidae, pp. 197-201. In C. Pozo [ed.], *Riqueza Biológica de Quintana Roo. Un Análisis para su Conservación. Tomo 2*, Colegio de la Frontera Sur, Conabio, Gobierno del Estado de Quintana Roo, México.
- Hernández-Baz, F. 2011b. Palomillas tigre (Insecta: Lepidoptera: Arctiidae), pp. 355-360. In A. Cruz Aragón [ed.], *La biodiversidad en Veracruz: Estudio de Estado. Vol. 2*, Conabio, Gobierno del estado de Veracruz, Universidad Veracruzana, Instituto de Ecología, A.C., México.
- Hernández-Baz, F. 2012. Biogeografía y conservación de las polillas avispa de México (Lepidoptera: Erebidae: Arctiinae Ctenuchina y Euchromiina). Editorial Académica Española, Saarbrücken, Deutschland.
- Hernández-Baz, F. 2013. Mariposas tigre (Arctiidae), pp. 66-70, 227-231. In A. A. Cruz, E. D. Melgarejo, F. Camacho, y K. C. Nálera [eds.], *La Biodiversidad de Chiapas: Estudio de Estado. Comisión para el Conocimiento y Uso de la Biodiversidad y Gobierno del estado de Chiapas*, México.
- Hernández-Baz, F. 2016. Mariposas nocturnas (Arctiidae), pp. 389-395. In A. Cruz, M. A. Ortega, E. D. Melgarejo, H. B. Perdoma, y A. V. Contreras. [coord.], *La Biodiversidad de Colima. Estudio de Estado*, Conabio, México.
- Hernández-Baz, F., and A. C. Bailey. 2006. Los ctenuchinae (Insecta: Lepidoptera: Arctiidae) de la República de Guatemala: una síntesis preliminar, pp. 403-413. In E. Cano [ed.], *La Biodiversidad de Guatemala*. Universidad del Valle de Guatemala, CONCYT, Guatemala.
- Hernández-Baz, F., J. M. González, and S. B. Vinson. 2012. Ecology and conservation of *Coreura albicosta* Draudt, 1916 (Lepidoptera: Erebidae: Arctiinae: Ctenuchina), an endemic species of Mexico. Southwest. Entomol. 37: 369-378.
- Hernández-Baz, F., R. Coates, J. A. Teston, and J. M. González. 2013. *Scena propylea* (Druce, 1894) (Lepidoptera: Erebidae: Arctiinae: Euchromiina) an endemic species of Mexico. Neotrop. Entomol. 42: 246-251.
- INEGI (Instituto Nacional de Geografía e Informática). 2012. Carta topográfica de México 1:250 000. Instituto Nacional de Estadística, Geografía e Informática, México. <http://mapserver.inegi.gob.mx/geografia/espanol/prodyser/cartas> Accessed 5 March 2022.
- Lafontaine, J. D., and M. Fibiger. 2006. Revised higher classification of the Noctuoidea (Lepidoptera). Can. Entomol. 138: 610-635.

- Lafontaine, J. D., and B. C. Schmidt. 2010. Annotated checklist of the Noctuoidea (Insecta, Lepidoptera) of North America north of Mexico. Zookeys 40: 1-239.
- Simmons, R. B., S. J. Weller, and S. J. Johnson. 2012. The evolution of androconia in mimetic tiger moths (Noctuoidea: Erebidae: Arctiinae: Ctenuchina and Euchromiina). Ann. Entomol. Soc. Am. 105: 804-816.

Natural History Notes of *Dasymutilla erythrina* (Hymenoptera: Mutillidae) in Biosphere Reserve Tehuacán-Cuicatlán, México

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Abstract. *Dasymutilla erythrina* (Say, 1836) (Mutillidae) as a visitor of *Ferocactus latispinus* (Britton & Rose, 1992) (Cactaceae) was reported in the Botanical Garden "Helia Bravo Hollis" Tehuacán-Cuicatlán Biosphere Reserve, Puebla, Mexico. General information about its natural history is included.

Resumen. Se reporta la presencia de *Dasymutilla erythrina* (Say, 1836) (Mutillidae) visitando flores de *Ferocactus latispinus* (Britton & Rose, 1992) (Cactaceae) en el Jardín Botánico "Helia Bravo Hollis" Reserva de la Biosfera Tehuacán-Cuicatlán, Puebla, México. Se adicionan datos sobre su historia Natural.

Velvet ants (Mutillidae) are distributed worldwide but are more abundant in tropical climates (Castro and Sesma 2010). The family has 216 valid genera and 30 valid subgenera, and about 8,000 species, but only about 4,300 of the species have been described (Lelej 2005, Lelej and Brothers 2008). Of all known species, 435 are in the South and West of North America (Triplehorn and Johnson 2005). Brothers and Lelej (2017) proposed nine monophyletic subfamilies. Three (Mutillinae, Myrmosinae, and Sphaerothalminae) are frequently found in the USA north of Mexico (Manley and Pitts 2002).

The genus *Dasymutilla* Ashmead (1899) has 139 neotropical species (Nonveiller 1990, Manley and Pitts 2007) found from the southern USA to northern South America (Cabra et al. 2018). The group is more diverse in arid and desert regions of its range (Manley and Tabers 1978). *Dasymutilla* is one of the richest genera and contains numerous species, but both sexes are known in only about a third of the species (Manley 2003, Pilgrim et al. 2008). Most species of the genus are diurnal, except *D. nocturna*, *D. paranocturna*, *D. arenivaga*, *D. megalophtalma*, and *D. subhyalina* that are nocturnal (Manley 1999).

Adult *Dasymutilla* are commonly "hairy" and frequently have contrasting colors (with areas of the body black, white, and/or red). Males feed on nectar, are winged,

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and similar in shape to many wasps, except their bodies are covered by dense coarse setae (Gardiner 2015). Females are apterous and their morphology is similar to that of ants (Brothers 2006). They are aposematic and protected by several secondary defenses, like stridulatory organs, pungent secretions, hard exoskeleton, and painful sting they use to warn or repel potential predators (Brach 1978).

The most important defense mechanism of velvet ants is the stinger formed by a modified ovipositor and, therefore, found only in females. The extremely painful sting of a related species, *D. klugii*, is rated 3 on a pain-level scale of 4 (Schmidt 2016). The stinger can be as long as the metasoma, almost half the length of the entire body (Manley 2000, Sadler et al. 2018). Although *Dasymutilla* spp. parasitizes mainly potentially important agricultural hosts (solitary bees and wasps), their defense methods probably evolved in response to pressure from predators (Wilson et al. 2012). The life cycles of velvet ants are almost completely unknown, and of the 139 species and subspecies of tropical and subtropical *Dasymutilla*, hosts of only 14 are known (Cabra et al. 2018).

The Botanical Garden "Helia Bravo Hollis" in Zapotitlán Salinas, Tehuacán, Puebla, Mexico, is part of the Tehuacán-Cuicatlán Biosphere Reserve (D.O.F. 2006). Vegetation of the region is typically xerophilous with abundance of cacti (Cactaceae) (Zavala-Hurtado 1982, Dávila-Aranda 1983). The Tehuacán-Cuicatlán Valley is an ecological island and contains a great diversity of flora and fauna, but also many isolated and endemic species (Rzedowski 1978, Arizmendi and Espinosa de los Monteros 1996, Morrone 2015, Halffter and Morrone 2017, García-Díaz et al. 2020, González et al. 2021). It has 2,700 species of vascular plants, and plant endemism is estimated at about 30% (Dávila-Aranda 1983). All cacti in the reserve are protected because they have some level of threat (SEMARNAT 2010). Many insects that visit flowers and extrafloral nectaries of Cactaceae of the region are little known, which is why we are reporting this velvet ant and its interaction with one of those plants.

The botanical garden "Helia Bravo Hollis" (Biosphere reserve Tehuacán-Cuicatlán) ($18^{\circ} 19'42.43''$ N / $97^{\circ}27' 16.94''$ W) at 1,446 m above sea level, in the Municipio de Zapotitlán Salinas, Puebla, México, was visited on 20 September 2018. While observing flowers of *Ferocactus latispinus* (Britton and Rose 1922) (Cactaceae) (Fig. 1A-C) at 1850 hours, a red-faced velvet ant, *D. erythrina*, was noted. The insect spun around the cactus plant and was "hiding" under bushes and thorns. The walking speed of the velvet ant was fast but gave the impression of being erratic, but the ant did not behave aggressively. The behavior seemed normal and had been reported previously (Brothers 2006).

The cacti flowers were observed from 0700 to 2000 hours for a single day. The cactus *F. latispinus* was identified based on Arias et al. (2000). The velvet ant was collected using ethyl acetate ($C_4H_8O_2$) and taken to a laboratory for identification. Based on Manley and Pitts (2002, 2007), we identified it as *Dasymutilla erythrina* (Say, 1836). Specimens were collected with a scientific license FAUT-0194 (SEMARNAT) of the first author. The material was temporarily deposited in the Lepidoptera Collection of the Faculty of Biology, Veracruzana University, Key: DF-CC-276-13 (SEMARNAT) Mexico.

Another specimen of *D. erythrina* captured in the same area a year earlier behaved in the same type of fast and erratic movement, making it difficult to capture. That specimen walked around a 100 m^2 area. The individual visited flowers of different cacti (unrecorded species), going in and out a few times in each. The specimen was collected at 1200 hours, a time of most insolation in the semi-desert region.



Fig. 1. (A) *Ferocactus lapispinus* (Britton & Rose, 1992) (Cactaceae) with open flower. Jardín Botánico “Helia Bravo Hollis,” Biosphere Reserve Tehuacán-Cuicatlán, Puebla, México. (B) *Dasymutilla erythrina* ♀ dorsal view. (C) *D. erythrina* ♀ ventral view. Photos: Fernando Hernández-Baz.

Material Examined. 1 ♀, México, Puebla, Zapotitlán Salinas, Jardín Botánico “Helia Bravo Hollis”, 1,447 m, 2018/09/18, 18°19' 51.47" N / 97° 27' 25.616" W. Colecta: 1800-1900 hours. Col. Sandra Lagunes Portugal and Fernando Hernández-Baz; 1 ♀, México, Puebla, Zapotitlán Salinas, Jardín Botánico “Helia Bravo Hollis”, 1,495 m, 2017/10/22, 18°19' 59.93" N / 97° 27' 30.19" W. Colecta: 1100-1200 hours. Col. Fernando Hernández-Baz.

Additional Comments. The Botanical Garden “Helia Bravo Hollis” at Valle de Zapotitlán Salinas, is characterized by dry, arid climate with scarce and irregular rain during the summer (Arias et al. 2000). Females of the velvet ant species are frequently found in sunny, sandy, and desertic areas (Joseph et al. 2012) and sometimes after sunset (Nonveiller 1963) that starts about 1800 hours in September. Although the two specimens mentioned were observed visiting cacti flowers, they were not observed feeding on nectar or pollen, or even on extrafloral nectaries as indicated by Elias (1983). Starting with this note, we intend to provide detailed analysis of distribution and behavior of velvet ants associated with the Biosphere Reserve of Tehuacán-Zapotitlán, México.

Acknowledgment

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Parasitología, code: UV-CA-421. Insects were collected with scientific license FAUT-0194 provided by Secretaría de Medio Ambiente Recursos Naturales, Mexico.

References Cited

- Arias, T., V. Valverde, and S. Reyes. 2000. Las plantas de la región de Zapotitlán Salinas, Puebla, SEMARNAT, Instituto Nacional de Ecología, Red para el Desarrollo Sostenible, A. C., UNAM, México, DF.
- Arizmendi, M. V. C., and A. Espinosa de los Monteros. 1996. Avifauna de los bosques de cactáceas columnares del Valle de Tehuacán, Puebla. Acta Zool. Mex., Nueva Serie 67: 25-46.
- Ashmead, W. H. 1899. Super-families in the Hymenoptera and generic synopses of the families Thynnidae, Myrmecidae, and Mutilidae. J. N. Y. Entomol. Soc. 7: 45-60.
- Brach, V. 1978. *Brachynemurus nebulosus* (Neuroptera: Myrmeleontidae): a possible Batesian mimic of Florida muddid wasps (Hymenoptera: Mutilidae). Entomol. News 89: 153-156.
- Brothers, D. J. 2006. Familia Mutilidae, pp. 577-594. In F. Fernández and M. J. Sharkey [eds.], Introducción a los Hymenoptera de la Región Neotropical. Sociedad Colombiana de Entomología, Universidad Nacional de Colombia; Bogotá, Distrito Capital, Colombia.
- Brothers, D. J., and A. S. Lelej. 2017. Phylogeny and higher classification of Mutilidae (Hymenoptera) based on morphological reanalysis. J. Hymenopt. Res. 60: 1-97. <https://doi.org/10.3897/jhr.60.20091>
- Cabra, R., K. Williams, D. Quintero, D. Windsor, J. Pickering, and D. Saavedra. 2018. *Dasymutilla* Ashmead (Hymenoptera: Mutilidae) in Panama: new species, sex associations and seasonal flight activity. Insecta Mundi 608: 1-17.
- Castro, L., and L. Sesma. 2010. Posible depredación de un nido de *Polistes biglumis* (Linnaeus 1758) por *Mutilla europea* Linnaeus 1758 (Hymenoptera, Vespidae; Mutilidae). BV News 2: 3-5.
- Dávila-Aranda, P. D. 1983. Flora genérica del Valle de Tehuacán-Cuicatlán. M.S. thesis, Universidad Nacional Autónoma de México, México, DF.
- D. O. F. (Diario Oficial de la Federación). 2006. Decreto por el que se declara área natural protegida, con el carácter de reserva de la biosfera, la región denominada Tehuacán-Cuicatlán, ubicada en los estados de Oaxaca y Puebla. Secretaría de Gobernación, México, DF. http://www.dof.gob.mx/nota_detalle.php?codigo=4893340&fecha=18/09/1998 Consulted 2 October 2021.
- Elias, T. S. 1983. Extrafloral nectaries: their structure and distribution, pp. 174-203. In B. L. Bentley and T. S. Elias [eds.], The Biology of Nectaries. Columbia University Press, New York.
- García-Díaz, J. J., J. Y. Miller, and J. M. González. 2020. Observations on the courtship and other biological aspects of *Athis hechiae* (Dyar, 1910) (Castniidae) in Tehuacán, Puebla, Mexico. Trop. Lepid. Res. 30: 86-89.
- Gardiner, M. 2015. Good Garden Bugs: Everything You Need to Know about Beneficial Insects. Quarry Books, China.
- González, J. M., B. Lopez Godinez, J. J. Garcia-Diaz, S. Simon, V. Sarto, I. Monteys, and R. Worthy. 2021. Reinstatement of *Athis miastagma* (Dyar, 1925) (Lepidoptera: Castniidae) as a valid species inhabiting the Pacific slope of Central Mexico. Zootaxa 5061: 300-322. <https://doi.org/10.11646/zootaxa.5061.2.4>

- Halfpter, G., and J. Morrone. 2017. An analytical review of Halfpter's Mexican transition zone, and its relevance for evolutionary biogeography, ecology, and biogeographical regionalization. Zootaxa 4226: 1-46. [PMid:28187628] <https://doi.org/10.11646/zootaxa.4226.1.1>
- Joseph, S., K. A. Wilson, M. L. Williams, C. D. Forister, and J. P. Von Dohlen. 2012. Repeated evolution in overlapping mimicry rings among North American velvet ants. Nat. Commun. 3: 1272. <https://doi.org/10.1038/ncomms2275>
- Lelej, A. S. 2005. Catalogue of the Mutillidae (Hymenoptera) of the Oriental Region. Dalnauka, Vladivostok.
- Lelej, A. S., and D. J. Brothers. 2008. The genus-group names of Mutillidae (Hymenoptera) and their type species, with a new genus, new name, new synonymies, new combinations and lectotypifications. Zootaxa 1889: 1-79.
- Manley, D. 1999. Synonymy of *Dasymutilla nocturna* Mickel (Hymenoptera: Mutillidae) Pan-Pac. Entomol. 75: 18-22.
- Manley, D. 2000. Defense adaptations in velvet ants (Hymenoptera: Mutillidae) and possible sources of selection pressure for such, pp. 285-289. In A. Austin and M. Dowton [eds.], Hymenoptera. Evolution, Biodiversity and Biological Control. CSIRO Publishing, Australia.
- Manley, D. 2003. *Dasymutilla jalisco*, a new species of velvet ant (Hymenoptera: Mutillidae) plus new synonymy for *Dasymutilla canina* (Smith). Proc. Entomol. Soc. Wash. 105: 679-684.
- Manley, D., and J. Pitts. 2002. A key to genera and subgenera of Mutillidae (Hymenoptera) in America North of Mexico with description of a new genus. J. Hymenopt. Res. 11: 72-100.
- Manley, D., and J. Pitts. 2007. Tropical and subtropical velvet ants of the genus *Dasymutilla* Ashmead (Hymenoptera: Mutillidae) with descriptions of 45 new species. Zootaxa 1487: 1-128.
- Manley, D., and S. Tabers. 1978. A mating aggregation of *Dasymutilla foxi* in southern Arizona (Hymenoptera: Mutillidae). Pan-Pac. Entomol. 54: 231-235.
- Nonveiller, G. 1963. Quelle est la cause de la rareté des Mutillides? Résultats de l'étude de certains de leur caractères biologiques et écologiques (Hymenoptera, Mutillidae). Memorie della Società Entomologica Italiana 42: 24-57.
- Nonveiller, G. 1990. Hymenopterorum Catalogus, Pars 18. SPB Academic Publishing, The Hague, Netherlands.
- Pilgrim, E. M., K. A. Williams, and J. P. Pitts. 2008. Sex association and synonymy in southwestern U. S. species of *Dasymutilla* (Hymenoptera: Mutillidae). Pan-Pac. Entomol. 84: 57-68.
- Rzedowski, J. 1978. Vegetación de México. Limusa, Ciudad de México.
- Sadler, E. A., J. P. Pitts, and J. S. Wilson. 2018. Stinging wasps (Hymenoptera: Aculeata), which species have the longest sting? PeerJ 6: e4743 <https://doi.org/10.7717/peerj.4743>
- Schmidt, J. O. 2016. The Sting of the Wild. John Hopkins University Press.
- SEMARNAT (Secretaría de Medio Ambiente y Recursos Naturales). 2010. Norma Oficial Mexicana NOM-059-2010, Protección ambiental-Especies nativas de México de flora y fauna silvestres-Categorías de riesgo y especificaciones para su inclusión, exclusión o cambio-Lista de especies en riesgo. Diario Oficial de la Federación 2010, México, DF.
- Triplehorn, C. A., and N. F. Johnson. 2005. Borror and DeLong's Introduction to the Study of Insects, 7th ed. Thomson Brooks/Cole, Belmont, CA.

- Wilson, J. S., K. A. Williams, M. L. Forister, C. D. Von Dohlen and J. P. Pitts. 2012. Repeated evolution in overlapping mimicry rings among North American velvet ants. *Nat. Commun.* 3: 1272.
- Zavala-Hurtado, J. 1982. Estudios ecológicos en el valle semiárido de Zapotitlán, Puebla. I. Clasificación numérica de la vegetación basada en atributos binarios de presencia o ausencia de las especies. *Biótica* 7: 99-120.

Eriophyid Mite as an Invasive Pest Associated with Gall Formation in Cotton from Five States of Mexico

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Abstract. Thirty accessions of cotton (*Gossypium*) in the INIFAP genebank in the Iguala Experimental Field were analyzed for resistance to eriophyid mites. Vegetative structures such as leaves and petioles and reproductive structures such as the bract calyx with galls were collected from cotton plants. Galls were cut into cross section to locate mites; taxonomic keys were used to identify the mites. Damage to accessions of cotton was evaluated on a scale of six classes. Damage by eriophyid mites was analyzed with PROC RANK, subjecting it to analysis of variance with PROC GLM, and mean comparison by Tukey ($P = 0.05$) using statistical software SAS® 9.2. Eriophyidae was reported associated with formation of galls in cotton. Severity of damage was 0 to 85%, but cotton accessions TOA-06, FZ-06, GOS-16, GOS-34, TOA-09, TOA-02, GOS-17, TOA-04, and TOA-05 were resistant. More studies of biology and ecology and molecular analysis are needed to better understand and develop management strategies against mites in cotton.

Introduction

Cotton (*Gossypium*), which belongs to the Malvaceae family, is a strategic crop for developing and developed countries (Solleiro 2016). It is the most used natural fiber in the world. Ninety-five percent of cotton produced in the world is *Gossypium hirsutum* L., with most wild populations in Mexico, a country considered the center of origin and domestication of cotton (Wegier 2013). During 2019, production was 25,315,342 tons mostly from Asia, America, and Africa, with India, China, and the United States the countries that produced the most. In Mexico, 368,000 tons were harvested (FAOSTAT 2022).

Ulloa et al. (2006) reported that Mexico is the center of origin of the genus *Gossypium*, with 11 of the 13-diploid species (*Gossypium armourianum* Kearney, *Gossypium lobatum* Gentry, *Gossypium gossypoides* (Ulbr.) Standl., *Gossypium aridum* (Rose & Standl.) Skovst., *Gossypium laxum* L.L. Phillips, *Gossypium shwendimanii* Fryxell & S.D. Koch, *Gossypium thurberi* Tod., *Gossypium trilobum* (DC.) Skovst., *Gossypium davisonii* Kellogg, *Gossypium turneri* Fryxell, and *Gossypium harknessii* Brandegee), and a tetraploid (*G. hirsutum*) comprising an important gene pool of the genus. Pérez et al. (2016) reported 980 accessions collected in Mexico from 1978-2006, with *G. hirsutum* the species most collected

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while least collected were *G. armourianum*, *G. shwendimanii*, *G. laxum*, *G. arboreum*, and *G. barbadense*.

The importance of a genebank of germplasm is availability of genes that could enable future ability to withstand new diseases, pests, or market demands by using conventional breeding or biotechnology to incorporate new genes into varieties. The Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP) at Iguala, Guerrero maintains a formal collection of cotton germplasm native to Mexico.

Production of good-quality, high yielding cotton allows Mexico to be competitive in the international market. But, since the beginning of the 1990s, production was affected by attacks from pests, with excessive use of agrochemicals resulting in environmental damage, high cost, and low profitability (SNICS 2020). Favorable international prices and use of genetically modified (GM) seed influence crop profitability. Use of GM seeds allows better pest control, less expenditure for insecticides, and greater yield despite being more expensive than conventional seeds (SNICS 2020, Gutiérrez 2021).

Estrada-Venegas et al. (2012) indicated the quarantine importance of mite pests in Mexico in which *Acalitus gossypii* Banks (Acarida: Eriophyidae) associated with wild cotton plants was recorded in an acarological collection for the country. *A. gossypii* has had other scientific names including *Aceria gossypii*, *Cecidophyes gossypii*, *Eriophyes gossypii*, and *Eryophyes gossypii* (CABI 2022). In India, *A. gossypii* associated with cotton was reported to cause 20-30% loss of yield (Rai et al. 1998). In accessions of native germplasm of INIFAP, gall-causing eriophyids are in vegetative and reproductive structures of cotton. The objective of this study was to analyze 30 accessions of cotton from the INIFAP genebank in Iguala Experimental Field for resistance to damage by eriophyid mites.

Materials and Methods

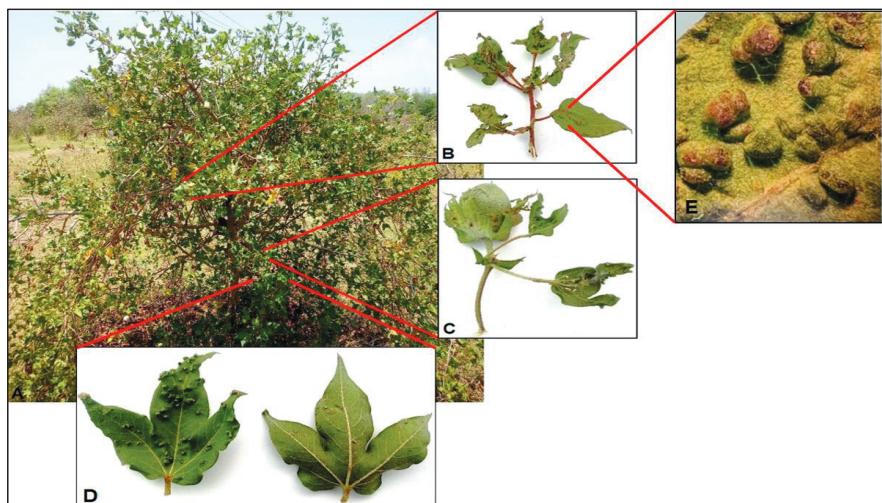


Fig. 1. Damage by eriophyid mites on cotton plants. A = complete cotton plant, B = new deformed leaves, leaf lamina and underside with galls (some deformed) and petioles with galls, C = leaf bract calyx with galls, D = lamina of leaf with galls and underside of leaf without galls, E = proximity of galls in leaf.

Vegetative and reproductive structures such as leaves, petioles, new outbreaks, and bract calyx with damage by mites (galls) (Fig. 1) were collected from 30 cotton accessions at Iguala de la Independencia, Guerrero, Mexico. The collection was established in 2019, and structures were collected in April 2022. The accessions are mostly *G. hirsutum* and to a lesser extent *G. barbadense* L. collected in five states in Mexico (Table 1). Galls were cut into cross section to identify mites to family by using taxonomic keys of Jeppson et al. (1975) and Amrine and Stasny (1994).

Table 1. Geographical Origin of Cotton Accessions from Five States of Mexico

Accession	Locality	Municipality	Region	State	Species
US-023	Vicente Guerrero	Venustiano Carranza	Los Llanos	Chiapas	<i>G. hirsutum</i>
US-027	Francisco Villa	Villa Flores	Frailesca	Chiapas	<i>G. hirsutum</i>
US-050	El Zapotal	San Pedro Pochutla	Costa	Oaxaca	<i>G. hirsutum</i>
US-051	Llano Grande Tonameca	San Pedro Pochutla	Costa	Oaxaca	<i>G. hirsutum</i>
US-055	La Barra del Potrero	Santa María Tonameca	Costa	Oaxaca	<i>G. hirsutum</i>
PM-15	Santiago Miahuatlán	Santiago Miahuatlán	Tehuacán	Puebla	<i>G. hirsutum</i>
FZ-06	Xochipala	Eduardo Neri	Norte	Guerrero	<i>G. hirsutum</i>
FZ-10	La Mojonera	Tepecuacuilco	Norte	Guerrero	<i>G. hirsutum</i>
FZ-13	Cuexcontlán	Tepecuacuilco	Norte	Guerrero	<i>G. hirsutum</i>
FZ-15	Huitzoco	Huitzoco	Norte	Guerrero	<i>G. hirsutum</i>
GOS-14	Santa Martha Chichihualtepec	Ejutla de Crespo	Valles Centrales	Oaxaca	<i>G. hirsutum</i>
GOS-15	Los Terrones	Coyuca de Catalán	Tierra Caliente	Guerrero	<i>G. hirsutum</i>
GOS-16	Tanganguato	Pungarabato	Tierra Caliente	Guerrero	<i>G. hirsutum</i>
GOS-17	Ixcateopan	Ixcateopan	Norte	Guerrero	<i>G. barbadense</i>
GOS-20	Sabanilla	Acapulco	Acapulco	Guerrero	<i>G. hirsutum</i>
GOS-27	Col. Centro	Marquelia	Costa Chica	Guerrero	<i>G. hirsutum</i>
GOS-34	Col. Miguel Alemán	Ometepec	Costa Chica	Guerrero	<i>G. hirsutum</i>
TOA-02	Puente rallado	Iguala de la independencia	Norte	Guerrero	<i>G. hirsutum</i>
TOA-03	Iguala	Iguala de la independencia	Norte	Guerrero	<i>G. hirsutum</i>
TOA-04	Casino de la Unión	Taxco de Alarcón	Norte	Guerrero	<i>G. hirsutum</i>
TOA-05	Teacalco	Amacuzac	Suroeste	Morelos	<i>G. hirsutum</i>
TOA-06	Teacalco	Amacuzac	Suroeste	Morelos	<i>G. hirsutum</i>
TOA-07	Ojo de agua	Amacuzac	Suroeste	Morelos	<i>G. hirsutum</i>
TOA-08	Barrio Santa Cruz	Izúcar de Matamoros	Mixteca	Puebla	<i>G. hirsutum</i>
TOA-09	Sección tercera	Tehuitzingo	Mixteca	Puebla	<i>G. hirsutum</i>
TOA-10	Tehuixtla	Tehuitzingo	Mixteca	Puebla	<i>G. hirsutum</i>
TOA-11	Cuaulutla	Tehuitzingo	Mixteca	Puebla	<i>G. hirsutum</i>
TOA-12	Sección primera	Tehuitzingo	Mixteca	Puebla	<i>G. hirsutum</i>
TOA-13	Sección primera	Tehuitzingo	Mixteca	Puebla	<i>G. hirsutum</i>
TOA-14	Sección primera	Tehuitzingo	Mixteca	Puebla	<i>G. hirsutum</i>

Damage to cotton was evaluated on a scale of six classes (Table 2). Cotton was in a randomized block design with three replications and each plant a replication. Damage by eriophyids was analyzed by PROC RANK, equivalent to a Kruskal-Wallis test in SAS® 9.2 (Shah and Madden 2004). Analysis of variance by PROC GLM was used to analyze damage. Tukey (SAS 2010) was used for mean separations among cotton accessions.

Table 2. Scale for Assessment of Damage by Eriophyid Mites in Cultivated Cotton

Class	% ^a	Description of damage
1*	0	Plant without galls
2	1-10	Plant with few galls, mostly only in lamina of leaves
3	11-30	Plant with few galls on lamina and underside of leaves
4	31-55	Plant with moderate galls on lamina and underside of leaves
5	56-85	Plant with most leaves covered by galls and some deformed leaves
6	>86	Plant with galls on lamina and underside of leaves, new outbreaks deformed, petioles and bract calyx with deformities

^aPercentage of damage by class. *Cotton plants resistant.

Results

Mites in galls from different cotton structures were Eriophyidae (Fig. 2), with a long, ringed, white-yellow body and two pairs of legs. Damage by Eriophyidae

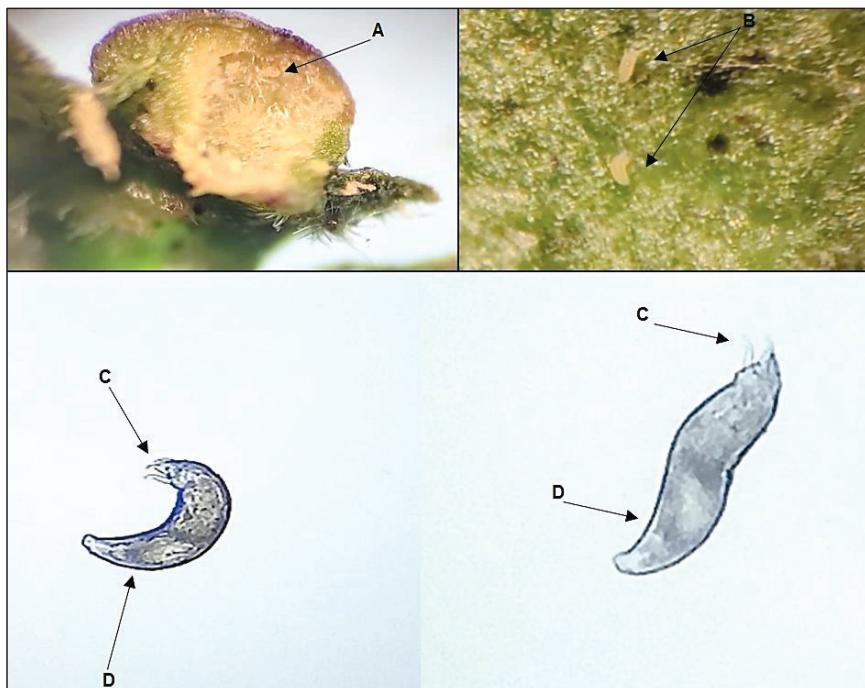


Fig. 2. Eriophyid mites from galls on plant structures. A = mites on inside of gall, B = mites on leaf surface, C = two pairs of legs, D = elongated and ringed body.

differed significantly among cotton accessions (Table 3) and ranged from 0 to 85% (Fig. 3). *G. hirsutum* (FZ-06, GOS-16, GOS-34, TOA-02, and TOA-04), *G. barbadense* (GOS-17) from the State of Guerrero, *G. hirsutum* (TOA-06 and TOA-05) from the State of Morelos, and *G. hirsutum* (TOA-09) from the State of Puebla were scored 1 for resistance.

Accessions of *G. hirsutum* most damaged by eriophyid mites (4 to 6 scale) with galls and 45-85% damage and scales of 1 to 3 with 7-27% damage and few to moderate galls in lamina and on the underside of leaves (broad and narrow and some deformed) were: TOA-08, US-023, PM-15, TOA-27, TOA-03, TOA-21, US-051, TOA-16, GOS-15, TOA-07, US-055, GOS-14, FZ-10, TOA-17, TOA-13, and GOS-13.

Table 3. Cotton Accessions Least Damaged by Eriophyid Mites, Iguala Experimental Field, 2022

Accession	Blade type	Rank-scale	Mean scale	Damage (%)
TOA-06	Narrow	6.833	c	1
FZ-06	Broad	6.833	c	1
GOS-16	Broad	6.833	c	1
GOS-34	Broad	6.833	c	1
TOA-09	Broad	6.833	c	1
TOA-02	Broad	6.833	c	1
GOS-17	Broad	6.833	c	1
TOA-04	Broad	6.833	c	1
TOA-05	Broad	6.833	c	1
GOS-13	Narrow	10.500	b c	1
TOA-13	Broad	11.667	b c	2
GOS-14	Narrow	12.333	a b c	2
FZ-10	Broad	12.333	a b c	2
TOA-17	Broad	12.333	a b c	2
TOA-16	Broad	17.833	a b c	2
GOS-15	Narrow	17.667	a b c	2
TOA-07	Broad	17.667	a b c	2
US-055	Broad	16.667	a b c	2
TOA-27	Narrow	20.000	a b c	3
TOA-03	Broad	20.000	a b c	3
TOA-21	Broad	20.000	a b c	3
US-051	Broad	18.833	a b c	3
TOA-08	Broad	22.333	a b c	3
US-023	Narrow	22.333	a b c	3
PM-15	Broad	22.000	a b c	3
TOA-11	Narrow	24.167	a b	4
US-050	Narrow	24.667	a b	4
FZ-15	Broad	24.500	a b	4
TOA-10	Narrow	27.000	a b	5
TOA-12	Broad	28.667	a	6

Mean damage to cotton leaf evaluated on scale of 1 to 6, where 1 = plant without gall (0% damage) to 6 = plant with galls on lamina and underside of leaves, new outbreaks, petioles, and bract calyx (>86% damaged). Scale values were analyzed by SAS rank (equivalent to Kruskal-Wallis) and ANOVA by Proc GLM. Tukey test after generating ranges set to $P \leq 0.05$.

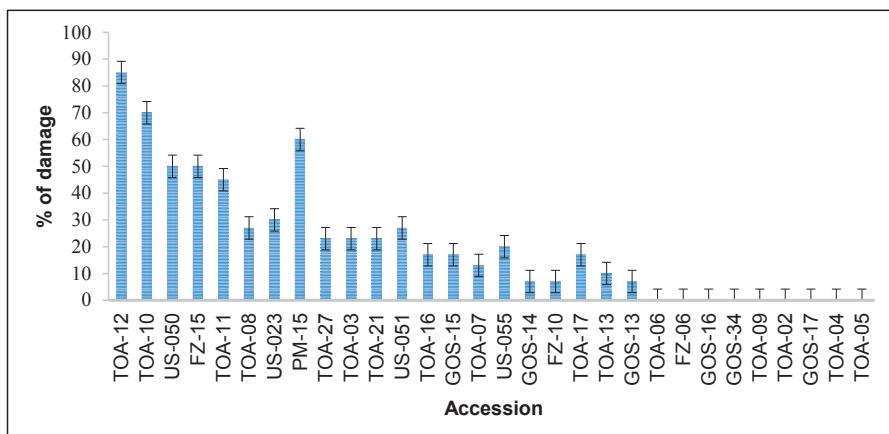


Fig. 3. Percentage of damage to cotton accessions collected in five Mexican states.

Discussion

Approximately 7,000 species of plant-damaging mites are known throughout the world (Krantz 2009), and identification of Eriophyidae was similar to that reported by several authors (Freeman et al. 2005, Howard et al. 2007, Domes 2009, Mielke-Ehret et al. 2010, Barraza 2014, Ferla et al. 2018, Ramírez et al. 2018, Trinidad et al. 2018). However, few species in the family worldwide are pests of crops (Smith et al. 2009). Eriophyidae was first reported on coconut palms (*Cocos nucifera* L., Arecaceae) by Keifer in 1965 at Guerrero, Mexico (Howard et al. 2007). Importance of mites as invasive species increased in recent years because the number of species displaced is increasing (Ferragut 2008). Phytophagous mites become adventitious pests in newly colonized areas because they and their potential to damage host plants are difficult to detect (Navia et al. 2007, 2010).

Eriophyid mites on the underside of leaves are protected from environmental factors (rain, sun, etc.), predators, parasitoids, and pesticides. In cotton accessions, the mites were in young and mature vegetative structures but damaged mostly immature structures.

Eriophyids prefer young leaves and have been studied for herbaceous attacking species such as *Aceria tosicella* Keifer and *Abacarus hystrix* (Nalepa) in wheat (*Triticum aestivum* L.) (Nault and Styer 1969, Gibson 1974, McCoy 1988, Davies and Albrigo 1994). Cotton with narrow (nine) and broad (21) leaves had galls by eriophyid mites, and some narrow leaves were deformed. Cristina (1999) alluded that leaves severely attacked by Eriophyidae and Tarsonemidae were deformed (from little to 25%). More leaves of cotton accessions in this study were deformed (Figs. 4-5), which affected longevity and photosynthetic ability.

Eriophyid mites show increasing ability to adapt to environmental factors, and each year specimens associated with new host plant species are found (Davis 2011, Aguilar and Murillo 2012, Bethke and Villavicencio 2014). This is important because they can be correlated with other pathogens. Arispe-Vázquez et al. (2021) mentioned that fall armyworm, *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae), was correlated with *Fusarium* spp. that causes ear rot of maize (*Zea mays* L., Poaceae).

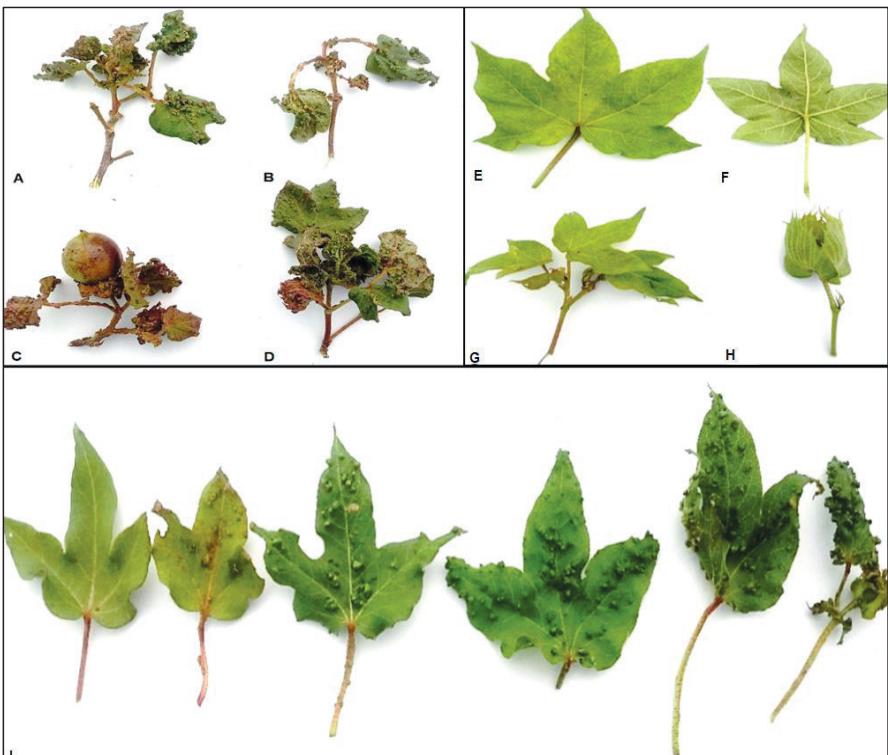


Fig. 4. Vegetative and reproductive structures of cotton accessions susceptible and resistant to damage by eriophyid mites. A = leaves and new outbreaks with galls (deformed leaves), B = lamina, underside of leaves and petioles with galls, C = leaves and bract calyx with galls, D = lamina and underside of leaves and new outbreaks with galls (very deformed leaves), E and F = lamina and underside of leaves without galls, G and H = new outbreaks and bract calyx without galls, I = leaves with different amounts of damage by mites.

Resistance is the ability of a plant to restrict, delay, or overcome infestation by a pest. A resistant plant yields more than a susceptible one when invaded by a pest (Badii and Garza-Almanza 2015). In this research, some cotton accessions were susceptible to damage by eriophyid mites, while others were more resilient and have potential to be used for genetic improvement. However, adequate weather, sufficient light, good water management, adequate nutrients, and integrated pest management are factors that also can influence resistance in crops (Koppert 2022).

In conclusion, Eriophyidae was reported associated with formation of galls in cotton accessions. The severity of damage by the family was 0 to 85%, but accessions TOA-06, FZ-06, GOS-16, GOS-34, TOA-09, TOA-02, GOS-17, TOA-04, and TOA-05 were resistant. More studies of the biology, ecology, and molecular analysis of the family are needed to better understand and develop management strategies in the cotton crop.



Fig. 5. Cotton accessions with narrow leaves with damage by eriophyid mites.

References Cited

- Aguilar, H., and P. Murillo. 2012. Nuevos hospederos y registros de ácaros fitófagos para Costa Rica: período 2008-2012. *Agronomía Costarricense* 36: 11-28.
- Amrine, J. W. Jr., and T. A. Stasny. 1994. Catalog of the Eriophyoidea (Acarina: Prostigmata) of the World. Indira Publishing House, West Bloomfield, Michigan, SA.
- Arispe-Vázquez, J. L., L. A. Aguirre-Uribe, E. Castro-del Ángel, Y. M. Ochoa-Fuentes, E. Cerna-Chávez, and A. Hernández-Juárez. 2021. Natural resistance of native and commercial maize to fall armyworm, *Spodoptera frugiperda*, and corn earworm, *Helicoverpa zea*, and their relationship with ear rot. *Southwest Entomol.* 46. doi.org:10.3958/059.046.0402
- Badii, M. H., and V. Garza-Almanza. 2015. Resistencia en Insectos, Plantas y Microorganismos. Cultura Científica Y Tecnológica 18. <http://erevistas.uacj.mx/ojs/index.php/culcyt/article/view/460>
- Barraza, A. E. 2014. Informe de visita técnica. BIOMUSEO, Amador, Panamá, República de Panamá.
- Bethke, J. A., and L. Villavicencio. 2014. Eriophyid mites on ornamental plants. Researchgate.net. University of California, Center for Applied Horticultural Research. 37-50.

- CABI (Centre for Agriculture and Bioscience International), online. Datasheet *Acalitus gossypii* (leaf blíster mite). Available online: <https://www.cabi.org/isc/datasheet/21853> [Accessed: 18 May 2022].
- Cristina, M. N. 1999. Ácaros de importancia agrícola en Colombia Rev. Fac. Nal. Agr. Medellín. 52: 321-363.
- Davies, F., and L. Albrigo. 1994. Citrus, pp. 172-178. In Jeff Atherson [ed.], CAB INTERNATIONAL.
- Davis, R. S. 2011. Eriophyid mites: bud, blister, gall and rust mite. UTAH PESTS, fact sheet. ENT-149-11, Sep. Utah State University.
- Domes, R. 2009. *Eriophyes rubin* sp. (Acarina, Eriophyidae) an *Rubus caesius* und *Rubus thyrsoideus*. Zeitschrift Für Angewandte Entomologie 46: 245-253. doi:10.1111/j.1439-0418.1960.tb01378.x
- Estrada-Venegas, E. G., J. A. Acuña-Soto, and M. Patricia. 2012. Relevancia de los ácaros como plagas de importancia cuarentenaria en México, pp. 1-60. En E. G. Estrada-Venegas, J. Acuña-Soto, M. P. Chaires-Grijalva, y A. Equihua-Martínez [eds.], Ácaros de Importancia Cuarentenaria en México: Presente, Pasado y Futuro. Sociedad Mexicana de Entomología, Colegio de Postgraduados, México.
- FAOSTAT (Food and Agriculture Organization Corporate Statistical Database). 2022. Crops and livestock products. <https://www.fao.org/faostat/en/#data/QCL/visualize>
- Ferla, N. J., D. E. Silva, D. Navia, J. M. Do Nascimento, L. Johann, and E. De Lillo. 2018. Occurrence of the quarantine mite pest *Aculus schlechtendali* (Acar: Eriophyidae) in apple orchards of Serra Gaúcha, Rio Grande do Sul state, Brazil. System. Appl. Acarol. 23: 1190. doi:10.11158/saa.23.6.14
- Ferragut, F. 2008. Impacto ecológico del ácaro invasor *Tetranychus evansi* en los cultivos hortícolas españoles. En Actas del VI Seminario Científico Internacional de Sanidad Vegetal, 22-26 Septiembre, La Habana, Cuba.
- Freeman, T. P., J. A. Goolsby, S. K. Ozman, and D. R. Nelson. 2005. An ultrastructural study of the relationship between the mite *Floracarus perrepae* Knihinicki & Boczek (Acariformes: Eriophyidae) and the fern *Lygodium microphyllum* (Lygodiaceae). Aust. J. Entomol. 44: 57-61. doi:10.1111/j.1440-6055.2005.00425.x
- Gibson, R. W. 1974. Studies on the feeding behaviour of the eriophyid mite *Abacarus hystricis*, a vector of grass viruses. Ann. Appl. Biol. 78: 213-217.
- Gutiérrez, H. M. 2021. Aspectos económicos de la producción de algodón en México. Universidad Autónoma de Chapingo. División de Ciencias Económicas-Administrativas. Tesis Doctoral.
- Howard, F. W., D. Moore, E. Abreu, and S. A. Gallo. 2007. Acaro del coco, *Aceria guerrerensis* Keifer (Arácnidae: Acari: Eriophyidae). EDIS. doi:10.32473/edis-in744-2007
- Jeppson, L. R., H. H. Keifer, and E. W. Baker. 1975. Mites injurious to economic plants. University of California Press, Berkeley.
- Koppert (Koppert México). 2022. Promueve el crecimiento y la resistencia de tus cultivos. <https://www.koppert.mx/retos/promueve-el-crecimiento-y-la-resistencia-de-tus-cultivos/>
- Krantz, G. W. 2009. Habits and habitats, pp. 64-82. In G. W. Krantz and D. E. Walter [eds.], A Manual of Acarology. Texas Tech University Press, Lubbock, TX.

- McCoy, C. W. 1988. Manejo de los principales insectos y ácaros de cítricos, pp. 29-37. En Miscelánea XV Sociedad Colombiana de Entomología 14. Socolen, Manizales.
- Mielke-Ehret, N., J. Thoma, N. Schlatermund, and H. P. Mühlbach. 2010. Detection of European mountain ash ringspot-associated virus-specific RNA and protein P3 in the pear leaf blister mite *Phytoptus pyri* (Eriophyidae). Archives of Virology 155: 987-991. doi:10.1007/s00705-010-0667-3
- Nault, R. L., and E. W. Styer. 1969. The dispersal of *Aceria tulipe* K. and three other grass-infesting eriophyd mites in Ohio. Ann. Entomol. Soc. Am. 62: 1446-1455.
- Navia, D., G. J. Moraes, and C. H. W. Flechtmann. 2007. Phytophagous mites as invasive alien species: quarantine procedures, pp. 307-316. In J. B. Morales-Malacara, V. Behan-Pelletier, E. Ueckermann, T. M. Perez, E. Estrada, C. Gispert, and M. Badii [eds.], Acarology: Proceedings of the XI international Congress, Merida, México.
- Navia, D., R. Ochoa, C. Welbourn, and F. Ferragut. 2010. Adventive eriophyoid mites: a global review of their impact, pathways, prevention and challenges. Exp. Appl. Acarol. 51: 225-255. <http://dx.doi.org/10.1007/s10493-009-9327-2>
- Pérez, M. C., G. Ma. R. Tovar, G. Q. Obispo, P. F. J. Legorreta, and C. J. A. Ruiz. 2016. Recursos genéticos del algodón en México: conservación ex situ, in situ y su utilización. REMEXCA 7: 5-16. <https://doi.org/10.29312/remexca.v7i1.366>
- Rai A.B, Patel J.R, Desai H.R, Patel A.J. Plant mites of agricultural importance in Gujarat. 252-261. In: National Seminar on Entomology in 21st Century organized by Entomological Society of India at Rajasthan College of Agriculture, Udaipur (Rajasthan) from April 30 to May 2, 1998.
- Ramírez, M. B., O. R. Arias, N. J. Zelada, V. A. Gómez, and G. J. de Moraes. 2018. Primer registro del ácaro fitófago *Dichopelmus notus* (Acari: Eriophyidae) en cultivo de yerba mate en la localidad de Tavaí, departamento de Caazapá, Paraguay. Investigación Agraria 20: 143-145. <https://dx.doi.org/10.18004/investig.agrar.2018.diciembre.143-145>
- SAS Institute. 2012. The SAS System for Windows, Release 9.1. SAS, Institute, Cary, NC.
- Shah, D.A., and L.V. Madden. 2004. Nonparametric analysis of ordinal data in designed factorial experiments. Phytopathology 94: 33-43.
- Smith, L., E. de Lillo, and J. W. Amrine. 2009. Effectiveness of eriophyid mites for biological control of weedy plants and challenges for future research. Exp. Appl. Acarol. 51: 115-149. doi:10.1007/s10493-009-9299-2
- SNICS (Servicio Nacional de Inspección y Certificación de Semillas). 2020. Programa de Abasto de Semilla de Algodón. SNICS, México. https://www.gob.mx/cms/uploads/attachment/file/632083/Programa_de_abasto_de_algodon_n.pdf
- Solleiro, R. J. L. 2016. Cadena de valor en la producción de algodón en México: los desafíos del mercado global. 21º Encuentro Nacional sobre Desarrollo Regional en México, 15-18 Noviembre 2016. AMECIDER - ITM, Mérida, Yucatán.
- Trinidad, C. T. O., M. E. Duarte, U. S. Da Cunha, and D. Navia. 2018. Eriophyoid mites associated with the blackberry in Brazil -- a new species in the genus *Diptacus* Keifer 1951 (Diptilomiopidae) and first report and supplementary

- description of *Acalitus orthomerus* (Keifer, 1951) (Eriophyidae). System. Appl. Acarol. 23: 1199. doi:10.11158/saa.23.6.15
- Ulloa, M.; Stewart, J. McD.; García, E. A. C.; Godoy, S. A.; Gaytán, A. M. and Acosta, S. N. 2006. Cotton genetic resources in the western states of México: in situ conservation status and germplasm collection for ex situ preservation. Genetic Res. Crop Evol. 53:653-668. <https://doi.org/10.1007/s10722-004-2988-0>
- Wegier, B. A. L. 2013. Diversidad genética y conservación de *Gossypium hirsutum* silvestre y cultivado en México. Tesis doctoral. Instituto de Ecología, Universidad Nacional Autónoma de México.

First Report of a Pest Mite on Medicinal Hemp Crops: Two-spotted Spider Mite, *Tetranychus urticae* Koch¹, at Azuay, Ecuador

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Abstract. Two-spotted spider mite, *Tetranychus urticae* Koch, was found infesting organic hemp crops at the municipality of Gualaceo-Azuay. In Ecuador, hemp recently is being cultivated for medical purposes. However, lack of information of the main pests related to the crop makes implementation of management strategies difficult. *Tetranychus urticae* is a phytophagous mite attacking many crops worldwide. The first report of the occurrence of the pest is important for public and private institutions involved in production of hemp to manage the mite. Future research should be focused on searching for an effective biological control agent for use in organic hemp crops where synthetic agrochemicals are not allowed.

Two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae), is a major pest of many crops worldwide (García-Marí and Enrique González-Zamora 1999, Skirvin and de Courcy Williams 1999, Greco et al. 2005, Oliveira et al. 2007, Vacacela Ajila et al. 2019). It is one of the most destructive pests of hemp in greenhouses and planting rooms (McPartland et al. 2000, Cranshaw et al. 2019). Light-colored spots on the upper part of the leaf indicate damage (McPartland et al. 2000). When *T. urticae* is abundant, leaves turn parched yellow, brown, and die (McPartland et al. 2000, Groves et al. 2020). The spider mite also produces webbing on leaves and in hemp flower buds (Britt et al. 2020).

In Ecuador, hemp cultivation is relatively new, so on 24 December 2019, the Organic Law Reforming the Integral Organic Criminal Code was published in the Official Registry (Asamblea Nacional República del Ecuador 2019). One of the most important aspects of this law is that it excludes non-psychoactive *Cannabis* (hemp) from narcotic and psychotropic substances with delta-9-tetrahydrocannabinol (THC) content less than 1%. Since then, hemp for medicinal purposes has started to be produced in some provinces of Ecuador (Pichincha, Azuay, Chimborazo). The first studies aimed to evaluate different varieties of the crop, but in other provinces, hemp already is produced commercially.

¹Acari: Tetranychidae

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In a greenhouse of the National Institute of Agricultural and Livestock Research (INIAP) at the municipality of Gualaceo (2.864978°S , $78.778301^{\circ}\text{W}$), Azuay, where hemp is produced, we found plants with typical symptoms caused by phytophagous mites: leaves with light-colored chlorotic spots (Fig. 1A-C). For 1 month (Mar 2022) we monitored hemp plants each week in the greenhouse. Each week, we collected leaves, put them into paper bags, and took them to the laboratory for additional analysis. The leaves were examined at 4x with the aid of a stereoscope (Olympus SZ51, Tokyo, Japan). We observed phytophagous mites in different stages of development (eggs, juveniles, and adults) on abaxial and adaxial leaf surfaces (Fig. 1D).



Fig. 1. Hemp plants damaged by two-spotted spider mite, *Tetranychus urticae* (A). Arrows indicate typical “light-colored chlorotic spots” in a greenhouse (B, C); individuals of *T. urticae* on the upper surface of a hemp leaf (D).

To identify species, individuals were collected and mounted in Hoyer medium on slides. Tetranychid males were mounted in a lateral position for examination of aedeagus ($n = 10$). Phase-contrast microscopy was used to aid in taxonomic identification. Diagnostic notes used for identification were those by Seeman and Beard (2011) for Tetranychidae. Specimens were kept in the Laboratory of Entomology at the National Institute of Agricultural and Livestock Research.

After analysis based on morphological characteristics, *T. urticae* was identified. Identification to species was based on the shape of the male aedeagus. The aedeagus had a small knob, pointed anterior and posterior projections of equal size, and convex dorsal surface (Seeman and Beard 2011) (Figs. 2A-C).

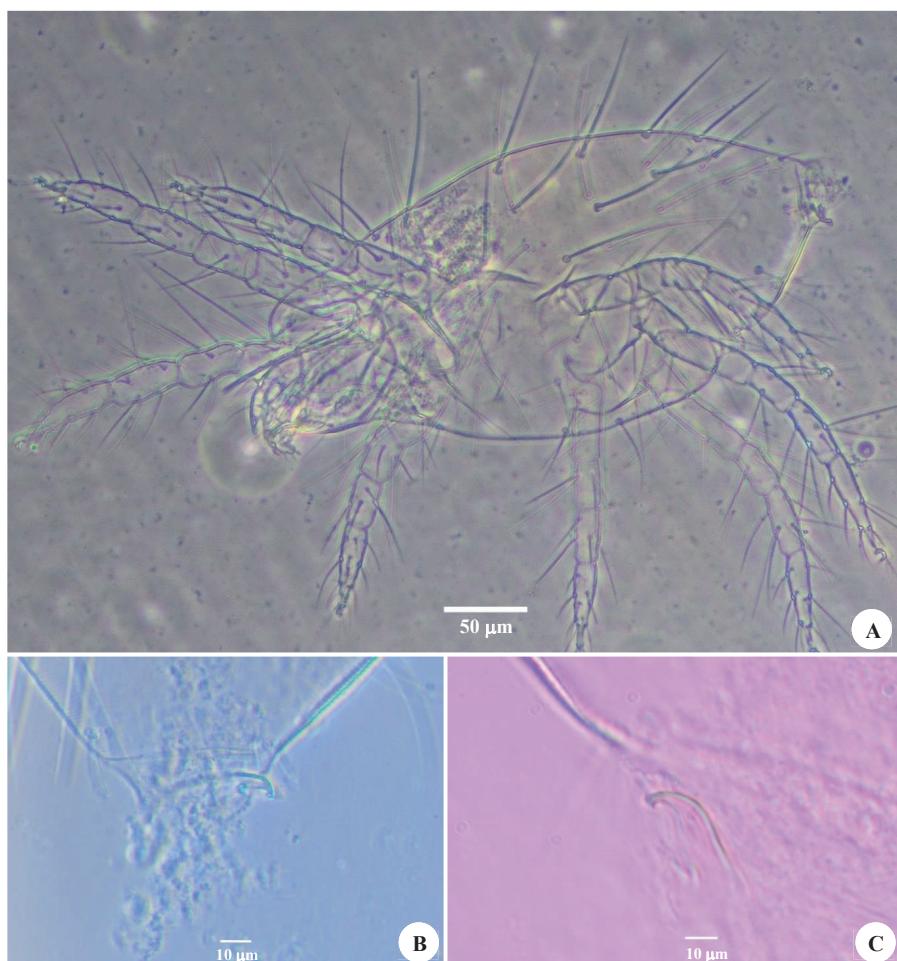


Fig. 2. Lateral view of a *Tetranychus urticae* male magnified 10 x with a microscope (A), and male aedeagus of *T. urticae* at 40x (B) and at 100x (C).

We reported the phytophagous two-spotted spider mite, *T. urticae*, infesting hemp plants at Azuay. We highlighted the importance of documenting the pest because the first hemp adaptability assays are being implemented in this region of Ecuador. Identification of pests on the crop is fundamental for future management. The severity of damage by this phytophagous mite is unquestionable because it is a major pest in different crops of the world (García-Marí and Enrique González-Zamora 1999, Skirvin and de Courcy Williams 1999, Greco et al. 2005, Oliveira et al. 2007, Vacacela Ajila et al. 2019). Several studies found the mite was resistant to different groups of acaricides (Carbonaro et al. 1986, Easterbrook et al. 2001). Because hemp was recently cultivated in Ecuador, no commercial acaricides are registered to control *T. urticae*. It is necessary to search or develop control methods compatible with organic farming. Biological control using predatory mites successful in other countries is a strategy that could be used to control *T. urticae* (García-Marí and Enrique González-Zamora 1999, Easterbrook et al. 2001, Opit et al. 2004, Oliveira et al. 2007). Future studies should verify the efficacy of the strategy and search for effective biological control agents.

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References Cited

- Asamblea Nacional República del Ecuador. 2019. Registro Oficial Nro. 107 Ley Orgánica Reformatoria al Código Orgánico Integral Penal.
- Britt, K., J. H. Fike, M. Flessner, C. Johnson, T. P. Kuhar, T. McCoy, and D. Reed. 2020. Integrated Pest Management of Hemp in Virginia. Virginia Cooperative Extension 29.
- Carbonaro, M. A., D. E. Moreland, V. E. Edge, N. Motoyama, G. C. Rock, and W. C. Dauterman. 1986. Studies on the mechanism of cyhexatin resistance in the twospotted spider mite, *Tetranychus urticae* (Acari: Tetranychidae). *J. Econ. Entomol.* 79: 576-579.
- Cranshaw, W., M. Schreiner, K. Britt, T. P. Kuhar, J. McPartland, and J. Grant. 2019. Developing insect pest management systems for hemp in the United States: a work in progress. *J. Integr. Pest Manage.* 10: 26.
- Easterbrook, M. A., J. D. Fitzgerald, and M. G. Solomon. 2001. Biological control of strawberry tarsonemid mite *Phytonemus pallidus* and two-spotted spider mite *Tetranychus urticae* on strawberry in the UK using species of *Neoseiulus* (*Amblyseius*) (Acari: Phytoseiidae). *Exp. Appl. Acarol.* 25: 25-36.
- García-Marí, F., and J. Enrique González-Zamora. 1999. Biological control of *Tetranychus urticae* (Acari: Tetranychidae) with naturally occurring predators in strawberry plantings in Valencia, Spain. *Exp. Appl. Acarol.* 23: 487-495.
- Greco, N. M., N. E. Sánchez, and G. G. Liljesthröm. 2005. *Neoseiulus californicus* (Acari: Phytoseiidae) as a potential control agent of *Tetranychus urticae* (Acari: Tetranychidae): effect of pest/predator ratio on pest abundance on strawberry. *Exp. Appl. Acarol.* 37: 57-66.
- Groves, R., P. Liesch, and B. Jensen. 2020. Insect and Mite Pests of Field Grown Hemp in Wisconsin. College of Agricultural and Life Sciences, University of Wisconsin-Madison 14.

- McPartland, J. M., R. C. Clarke, and D. P. Watson. 2000. Hemp Diseases and Pests: Management and Biological Control. CABI.
- Oliveira, H., A. Janssen, A. Pallini, M. Venzon, M. Fadini, and V. Duarte. 2007. A phytoseiid predator from the tropics as potential biological control agent for the spider mite *Tetranychus urticae* Koch (Acari: Tetranychidae). Biological Control 42: 105-109.
- Opit, G. P., J. R. Nechools, and D. C. Margolies. 2004. Biological control of twospotted spider mites, *Tetranychus urticae* Koch (Acari: Tetranychidae), using *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae) on ivy geranium: assessment of predator release ratios. Biological Control 29: 445-452.
- Seeman, O. D., and J. J. Beard. 2011. Identification of exotic pest and Australian native and naturalised species of *Tetranychus* (Acari: Tetranychidae). Zootaxa 2961: 1-72.
- Skirvin, D. J., and M. de Courcy Williams. 1999. Differential effects of plant species on a mite pest (*Tetranychus urticae*) and its predator (*Phytoseiulus persimilis*): implications for biological control. Exp. Appl. Acarol. 23: 497-512.
- Vacacela Ajila, H. E., F. Colares, F. Lemos, P. H. Marques, E. C. Franklin, W. Santos do Vale, E. E. Oliveira, M. Venzon, and A. Pallini. 2019. Supplementary food for *Neoseiulus californicus* boosts biological control of *Tetranychus urticae* on strawberry. Pest Management Science 75: 1986-1992.

New Records of Oribatid Mites (Acari: Oribatida) from Central Mexico

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Abstract. Nine species of oribatid mites in nine genera and nine families were reported for the first time from Central Mexico: *Arphthicularus gyros* Niedbała, 2004, *Nanhermannia elegantissima* Hammer, 1958, *Camisia khencensis* Hammer, 1961, *Epiereimulus geometricus* (Berlese, 1916), *Peloribates (P.) muscicola* Hammer, 1961, *Lucoppia burrowsi* (Michael, 1890), *Scheloribates (Bischeloribates) bidactylus* Hammer, 1961, *Parapirnodus prosopis* Martínez, Fernández and Monetti, 1996, and *Pergalumna (P.) ventralis* (Willmann, 1932). This work increased the number of oribatid mite records for Queretaro (eight species), Michoacan (13 species), and Mexico City (15 species).

Introduction

Oribatida is a diverse order of mites in the soil, primarily in natural ecosystems, where they are important for recycling nutrients (Norton and Behan-Pelletier 2009). Mexico is the country with the second largest number of species recorded in Latin America, surpassed only by Brazil (Oliveira et al. 2017).

Palacios-Vargas and Iglesias (2004) reported slightly more than 440 species in 250 genera of 104 families. Quintana Roo, Veracruz, Campeche, and Hidalgo are the states with most records in Mexico. However, in recent years important contributions have been made to knowledge of the group, which leads to speculation that the number will increase considerably (Bernal et al. 2009, Palacios-Vargas et al. 2011, Vázquez et al. 2016, Villagomez y Palacios-Vargas 2017, Ojeda and Gasca-Pineda 2019, Villagomez et al. 2019, Ojeda et al. 2020). Nine species of oribatid mites were reported for the first time from Central Mexico.

Materials and Methods

Four sites were sampled in the study: Site 1: avocado orchard with relicts of *Pinus michoacana* Martínez forest at Zumpimoto, State of Michoacán ($19^{\circ}21'07.2\text{ "N}$ $102^{\circ}04'02.7\text{ "W}$), 1,547 m above sea level; Site 2: natural area principally covered by *Bursera simaruba* Sarg. (1890) and scattered shrubs, Hotel la Misión, Concá, ($21^{\circ}25'50.5\text{ "N}$ $99^{\circ}36'44.3\text{ "W}$), 568 m above sea level and Site 3: *Quercus* spp. forest, rocky soil of volcanic origin with evidence of grazing, El Sabinito, Conca ($21^{\circ}26'57.3\text{ "N}$ $99^{\circ}37'00.5\text{ "W}$), 707 m above sea level in the State of Querétaro; and Site 4: forest of *Abies religiosa* (Kunth) Schltdl. and Cham., Desierto de los Leones,

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Mexico City, ($19^{\circ}18'20.4\text{ "N}$ $99^{\circ}18'11.8\text{ "W}$), 2,342 m above sea level (Fig. 1A). Five soil samples (15 cm^3) were collected from each ecosystem and processed in Berlese-Tullgren funnels to extract oribatid mites. From Sites 1 and 3, five samples of foliage also were collected and washed with soap-and-water solution and filtered in a $400\text{-}\mu\text{m}$ sieve to extract mites; *B. simaruba* trunks were collected from Site 2 and put into an emergence chamber and checked daily. Individuals were counted, macerated in lactic acid, and mounted in Hoyer's medium (Walter and Krantz 2009). The material was deposited in the private collection of acarology of the ENTOMOACARI laboratory at La Trinidad, Texcoco, State of Mexico, Mexico. Schatz et al. (2011) was used for classification to superfamily and family, and nomenclature for genera and species, biogeographic distributions, and synonymies were obtained from Subías (2004 update 2022).

Live specimens were photographed with the aid of a Canon Rebel T6® camera adapted to an AmScope® SM-2TZZ stereomicroscope. The slides were studied with the aid of a Carl Zeiss® Axioskop 2 plus phase-contrast microscope. Method B (depth map) of the Helicon Focus 7® program was used for rendering. Final editing was with GIMP version 2.10.20.

Results and Discussion

Nine species of oribatid mites in nine genera of nine families were reported for the first time for Mexico; four of the species were in the superfamily Oripodoidea.

Phthiracaridae: Phthiracaroidea

Arphthicarus gyros Niedbała, 2004 (Fig. 1B).

Material examined: 1 ♂, on *Quercus* spp. forest floor at Site 3.

World distribution: Guyana [British Guayana].

Observations: of the genus *Hoplophorella* only *A. sororius* (Niedbała, 1982) have been reported for the country (Balogh and Balogh 1988).

Nanhermanniidae: Crotonioidea

Nanhermanniaelegantissima Hammer, 1958 (Fig. 1C)

Material examined: 1 ♀, on *Quercus* spp. forest floor at Site 3.

World distribution: Neotropical and sub-Antarctic: I. Malvinas [I. Falkland]

Observations: the genus *Nanhermannia*, characterized by a rounded posterior part of the notogaster, lacking elongated tubercles and long notogastral setae; reported for the states of Quintana Roo and Oaxaca, however, so far there are no registered species (Bernal et al. 2009, Vázquez et al. 2016). Species of the family Nanhermanniidae have also been reported in genera *Cyrthermannia* Balogh, 1958 and *Masthermannia* Berlese, 1913 (Vázquez and Prieto 2001).

Crotoniidae: Crotonioidea

Camisia khencensis Hammer, 1961 (Fig. 1D)

Material examined: 12 ♀, 3 immatures (2 protonymphs and 1 tritonymph), on *Abies religiosa* foliage from Site 4.

World distribution: Neotropical (Peru and Chile).

Observations: Palacios-Vargas et al. (2003) and Villagomez et al. (2019) report *C. ca. segnis* and *C. cf. horrida*, respectively, for the country.

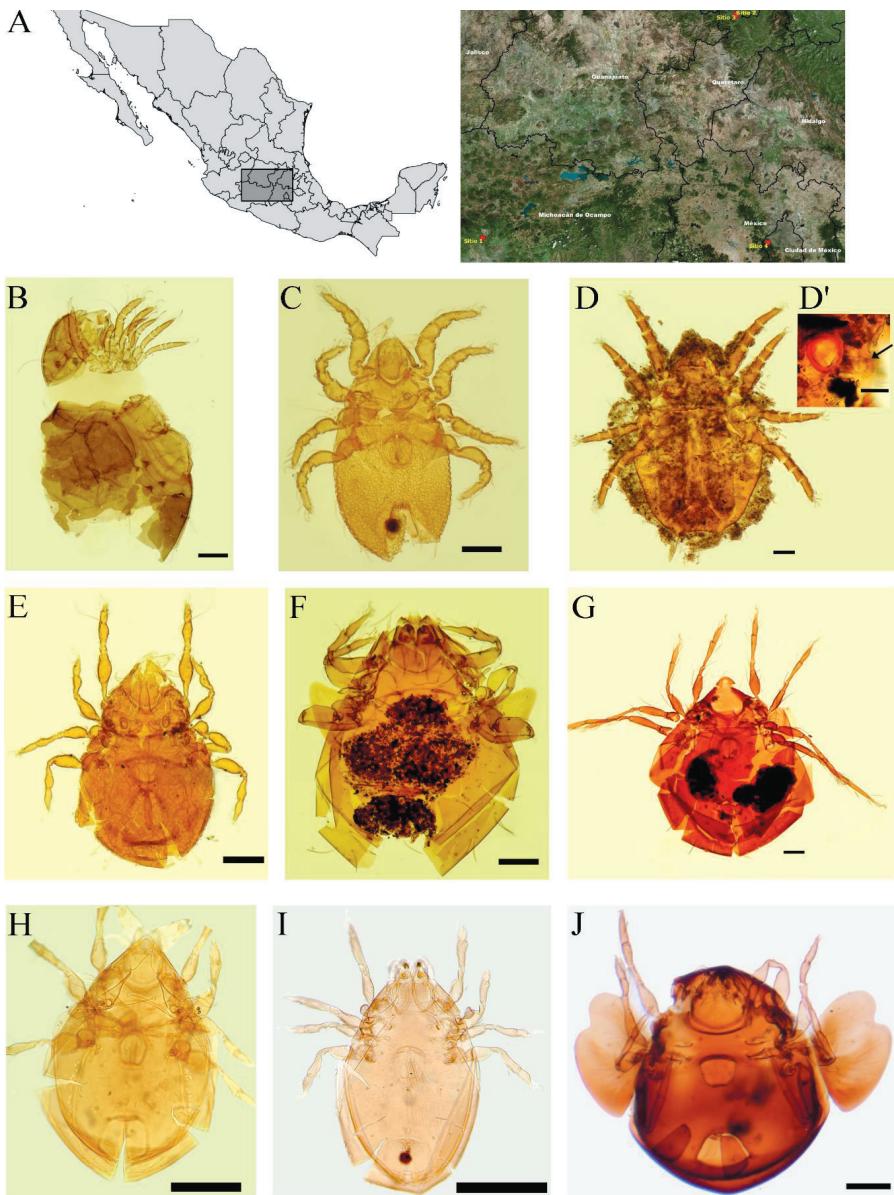


Fig. 1. A. Location of sampling sites. B. *Arphthicularus gyros* Niedbała, 2004 (♂). C. *Nanhermannia elegantissima* Hammer, 1958 (♀). D. *Camisia khencensis* Hammer, 1961 (♀), D'. Sensillum. E. *Epieremulus geometricus* (Berlese, 1916) (♂). F. *Peloribates (P.) muscicola* Hammer, 1961 (♂). G. *Lucoppia burrowsi* (Michael, 1890) (♂). H. *Scheloribates (Bischeloribates) bidactylus* Hammer, 1961 (♀). I. *Parapimnodus prosopis* Martínez, Fernández and Monetti, 1996 (Fig. 1I). J. *Pergalumna (P.) ventralis* (Willmann, 1932) (♀). Scale bar: 100 µm (A-J), 25 µm (D').

Caleremaeidae: Ameroidea

***Epipterymus geometricus* (Berlese, 1916)** (Fig. 1E)

=*Carabodooides saccharomyctoides* Jacot, 1937

Material examined: 1 ♂, in *Pinus michoacana* leaf litter from Site 1.

World distribution: United States (Eastern)

Observations: Vázquez (1999) and Vázquez et al. (2016) report to *E. braziliensis* (Balogh and Mahunka, 1969) and *E. granulatus* (Balogh and Mahunka, 1979), with material collected in the State of Quintana Roo.

Haplozetidae: Oripodoidea

***Peloribates (P.) muscicola* Hammer, 1961** (Fig. 1F).

Material examined: 6 ♀ y 11 ♂, in *Pinus michoacana* leaf litter from Site 1.

World distribution: Peru and Japan

Observations in the country have also been reported *P. (P.) grandis* (Willmann, 1930), *P. (P.) genavensium* Mahunka, 1983, *P. (P.) robustus* Grishina, 1981, *P. (Aokibates) yoshii* (Mahunka, 1988) and *P. (Peloribatodes) europaeus* Willmann, 1935 (Balogh and Balogh 1990; Vázquez et al. 2011, 2016; Subías 2022).

Oribatulidae: Oripodoidea

***Lucoppia burrowsi* (Michael, 1890)** (Fig. 1G).

Material examined: 1 ♂, in *Pinus michoacana* foliage from Site 1.

=*Zygoribatula apletosa* Higgins et Woolley, 1975

=*Phauloppia banksi* Marshall, Reeves and Norton, 1987

=*Zygoribatula longisetosa* Golosova, 1970

=*Romanobates maiensis* Choi, 1995

=*Lucoppia orientalis* Djaparidze, 1985

=*Notaspis pectinata* Hall, 1912, nam. praeoc. by Michael, 1885

=*Scutovertex pilosus* Banks, 1895

=*Eremaeus sanremensis* Oudemans, 1900

=*Zygoribatula spinosissima* Mihelčič, 1956

World distribution: Holarctic (western Palearctic: less frequent in the North, eastern Palearctic and Nearctic) and tropical (southeastern China and Hawaii).

Observations: *L. feideri* Subías, 2010 was previously reported in the country, from nest samples of *Aquila chrysaetos canadensis* Linnaeus, 1758 (Falconiformes: Accipitridae) collected in the State of Aguascalientes (Damián et al. 2019).

Scheloribatidae: Oripodoidea

***Scheloribates (Bischeloribates) bidactylus* Hammer, 1961** (Fig. 1H)

Material examined: 9 ♀ y 5 ♂, in trunks of *Bursera simaruba* from Site 2.

World distribution: Neotropical

Observations: Currently 14 species of the genus *Scheloribates* have been reported in Mexico (Vázquez 1999; Palacios-Vargas et al. 2011; Vázquez et al. 2011, 2016; Villagomez and Palacios-Vargas 2017; Villagomez et al. 2019; Revelo-Tobar et al. 2021).

Oripodidae: Oripodoidea

***Parapirnodus prosopis* Martínez, Fernández y Monetti, 1996** (Fig. 1I)

Material examined: 3 ♀ y 5 ♂, in *Quercus* spp. foliage at Site 3.

World distribution: Argentina

Observations: *Parapirnodus* is a genus with only three species. Palacios-Vargas et al. (2003) report to *P. ca. longus* and Woolley (1971) describes *P. denaius* with specimens fossilized in amber from Simojovel, Chiapas.

Galumnidae: Galumnoidea

***Pergalumna (P.) ventralis* (Willmann, 1932)** (Fig. 1J).

=*Galumna duplicita* Hammer, 1958

Material examined: 2 ♀, in *Quercus* spp. forest floor at Site 3.

World distribution: Tropical (Oriental, Polynesian and Neotropical) and Holarctic (U.S.A. and Palearctic: Eastern Mediterranean and East of Eastern Palearctic).

Observations: eight species of the genus *Pergalumna* were reported in the country, with specimens obtained from samples collected principally in the states of Veracruz and Quintana Roo (Vázquez et al. 2016, Villagomez and Palacios-Vargas 2017, Páez et al. 2019, Villagomez et al. 2019).

Although diversity, abundance, and importance of oribatids in the soil are unquestionable, there is great diversity of species in the canopy, such as those belonging to the genera *Camisia*, *Parapirnodus*, and *Scheloribates*. They fulfill various functions such as stimulating growth of certain algae and lichens, and their fecal pellets are a source of nutrients important for various species of arboreal microflora (Jensen 1974, Prinzing and Wirtz 1997). Many tree-canopy oribatids underwent various physiological, reproductive behavioral, and morphological changes (increased sexual dimorphism, short sensillum with globose or mass-shaped heads, enlarged pore areas, expanded solenidia, mono- or bi-dactyl claws and saccules on the legs) (Behan-Pelletier and Walter 2009). The features are consistent in several of the arboreal species reported in this manuscript: *P. prosopis* and *S. (S.) bidactylus* have mono- and bi-dactyl claws, respectively; *C. khencensis* has an elongate sensillum, with a rounded, flattened head, with an inner bubble and tubercles on it. Tubercles can be difficult to observe because of much organic matter the species usually carries in its body, and this would explain differences in interpretation as seen in diagrams of their description and redescription (Hammer 1961, Colloff 1993) (Fig. 1D). *L. burrowsi* shows evident porous areas, principally Aa. that is elongated and arranged longitudinally on the notogaster. This contribution increased the number of records of oribatid mite species in the states of Queretaro (eight species), Michoacan (25 species), and Mexico City (15 species) (Palacios-Vargas and Iglesias 2004, Sánchez-Rocha 2014, Revelo-Tobar et al. 2021, 2022).

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References Cited

- Balogh, J., and P. Balogh. 1988. The soil mites of the world. Oribatid mites on Neotropical Region 1. Akadémiai Kiado.
- Balogh, J., and P. Balogh. 1990. The soil mites of the world. Oribatid mites on Neotropical Region II. Akadémiai Kiado.
- Behan-Pelletier, V., and D. E. Walter. 2009. Biodiversity of oribatid mites (Acar: Oribatida) in tree canopies and litter, pp. 187-202. In D. Coleman and P. F.

- Hendrix [eds.], Invertebrates as Webmasters in Ecosystems. CABI Publishing. <https://doi.org/10.1079/9780851993942.0187>
- Bernal, A., G. Castaño-Meneses, J. G. Palacios-Vargas, and N. García. 2009. Oribatid mites and springtails from a coffee plantation in Sierra Sur, Oaxaca, Mexico. *Pesquisa Agropecuária Brasileira* 44: 988-995. <https://doi.org/10.1590/s0100-204x2009000800030>
- Colloff, M. J. 1993. A taxonomic revision of the oribatid mite genus *Camisia* (Acar: Oribatida). *Journal Natural History* 1325-1428.
- Damián, M., G. Villegas, and L. Lozano. 2019. Ácaros asociados a nidos de águila real (*Aquila chrysaetos canadensis*) (Falconiformes: Accipitridae) en México. *Acta Zool. Mex. (n.s.)* 35: 1-16. <https://doi.org/10.21829/azm.2019.3502196>
- Hammer, M. 1961. Investigations on the oribatid fauna of the Andes Mountains II, Perú. *Biologiske Skrifter Udgivet Af Det Kongelige Danske Videnskabernes Selskab* 13: 1-207.
- Jensen, V. 1974. Decomposition of angiosperm tree leaf litter, pp. 69-104. In D. Dickinson and G. Pugh [eds.], *Biology of Plant Litter Decomposition*. Vols. 1 and 2. Academic Press.
- Norton, R. A., and V. Behan-Pelletier. 2009. Suborder Oribatida, pp. 430-564. In G. W. Krantz and D. E. Walter [eds.], *A Manual of Acarology*. Texas Tech University Press.
- Ojeda, M., and J. Gasca-Pineda. 2019. Abundance and diversity of the soil microarthropod fauna from the Cuatro Ciénelas Basin, pp. 29-51. In F. Álvarez y M. Ojeda [eds.], *Animal Diversity and Biogeography of the Cuatro Ciénelas Basin*. https://doi.org/10.1007/978-3-030-11262-2_3
- Ojeda, M., P. Velez, L. Espinosa-Asuar, L. E. Eguiarte, and V. Souza. 2020. A new species of *Trhypochthoniellus* (Acari: Oribatida: Trhypochthoniidae) from Cuatro Ciénelas, Coahuila, Mexico, and a key to the world species. *Systematic and Applied Acarology* 25: 974-985. <https://doi.org/10.11158/saa.25.6.3>
- Oliveira, A. R., P. S. Argolo, G. J. De Moraes, R. A. Norton, and H. Schatz. 2017. A checklist of the oribatid mite species (Acar: Oribatida) of Brazil. *Zootaxa* 4245: 1-89. <https://doi.org/10.11646/zootaxa.4245.1.1>
- Páez, J., F. Villagomez, and J. G. Palacios-Vargas. 2019. Description of a new *Pergalumna* (Acari: Oribatida: Galumnidae) species from Mexico and its postembryonic development. *Zootaxa* 4647: 385-406. <https://doi.org/10.11646/zootaxa.4647.1.25>
- Palacios-Vargas, J. G., and R. Iglesias. 2004. *Oribatei* (Acari), pp. 431-468. In J. Llorente Bousquets, J. Morrone Juan, O. Yáñez Ordóñez, and I. Vargas Fernández [eds.], *Biodiversidad, Taxonomía y Biogeografía de Artrópodos: Hacia una Síntesis de su Conocimiento*. CONABIO.
- Palacios-Vargas, J. G., R. Iglesias, and G. Castaño-Meneses. 2003. Mites from Mexican oak canopies. *Int. J..Trop. Insect Sci.* 23: 287-292. <https://doi.org/10.1017/s1742758400012340>
- Palacios-Vargas, J. G., G. Castaño-Meneses, and D. Estrada. 2011. Diversity and dynamics of microarthropods from different biotopes of Las Sardinas Cave (Mexico). *Subterranean Biology* 9: 113-126. <https://doi.org/10.3897/subtbiol.9.2514>

- Prinzing, A., and H. Wirtz. 1997. The epiphytic lichen, *Evernia prunastri* L., as a habitat for arthropods: shelter from desiccation, food limitation and indirect mutualism, pp. 477-494. In N. E. Stork, J. Adis, and R. K. Didham [eds.], *Canopy Arthropods*. Chapman and Hall, London.
- Revelo-Tobar, H., Estrada-Venegas, E. G., Equihua-Martinez, A. and Valdez-Carrasco, J. 2021. New records of oribatid mites from Michoacán state, Mexico. *Entomological Communications* 3: 1-4. <https://doi.org/10.37486/2675-1305.ec 03049>
- Revelo-Tobar, H., Estrada-Venegas, E. G. and Equihua-Martinez, A. New records of oribatid mites from Mexico with description of a new species. *Zootaxa* 5168 (1): 024–038. <https://doi.org/10.11646/zootaxa.5168.1.2>
- Sánchez-Rocha, I. 2014. Algunos aspectos ecológicos de los ácaros del suelo de la cuenca de río Estórax, Qro. UNAM.
- Schatz, H., V. Behan-Pelletier, B. OConnor, and R., A. Norton. 2011. Suborder Oribatida van der Hammen, 1968, pp. 3148, 141-148. In Z. Zhang [ed.], *Animal Biodiversity: An Outline of Higher-level Classification and Survey of Taxonomic Richness*. Zootaxa.
- Subías, L. 2004. Listado sistemático, sinonímico y biogeográfico de los ácaros oribátidos (Acariformes: Oribatida) del mundo (1758-2002). *Graellsia* 60: 3-305.
- Subías, L. 2022. Listado sistemático, sinonímico y biogeográfico de los Ácaros Oribátidos (Acariformes: Oribatida) del mundo (excepto fósiles), 17^a actualización. http://bba.bioucm.es/cont/docs/RO_1.pdf
- Vázquez, M. 1999. Catálogo de los Ácaros Oribátidos Edáficos de Sian Ka'an. 1 ed. Universidad de Quintana Roo, Q. Roo, México.
- Vázquez, M., and D. Prieto. 2001. Oribatida, pp. 71-84. In *Fauna Edáfica de las Selvas Tropicales de Quintana Roo*. Universidad de Quintana Roo.
- Vázquez, M., A. Pereira, P. Fragoso, y J. Rodríguez. 2011. Riqueza y biodiversidad de organismos edáficos del Corredor Biológico Mesoamericano (México, Belice y Guatemala), pp. 37-70. In *Avances Deficiencia y Tecnología en Quintana Roo*.
- Vázquez, M., D. May, and E. Alamilla. 2016. Riqueza específica y biodiversidad de Cozumel, Quintana Roo, México. *Teoría y Praxis* 12: 137-171. <https://doi.org/10.22403/uqroomx/typ19/07>
- Villagomez, F., and J. G. Palacios-Vargas. 2017. Los ácaros oribátidos (acari: oribatei) de los estratos edáficos y arbóreos de la selva alta perennifolia de los Tuxtlas, Veracruz. *Entomología Mexicana* 4: 28-34.
- Villagomez, F., J. G. Palacios-Vargas, G. Castaño-Meneses, and I. Castellanos-Vargas. 2019. Effect of tree identity, temporal variation and edaphic parameters on the structure of the edaphic community of oribatid mites in an evergreen tropical forest of Mexico. *Applied Ecology and Environmental Research* 17: 14621-14639. https://doi.org/10.15666/aeer/1706_1462114639
- Walter, D. E., and G. W. Krantz. 2009. Collecting, rearing, and preparing specimens, pp. 83-96. In G. W. Krantz and D. E. Walter [eds.], *A Manual of Acarology*. Texas Tech University Press.
- Woolley, T. 1971. Studies of fossiliferous amber arthropods of Chiapas, Mexico, p. 10. In A. Petrunkevitch [ed.], *Fossiliferous Amber Arthropods*. University of California Press.

***Pseudophilothrips perseae*¹ Damaging Hass Avocado Fruits in México**

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Abstract. During 2020, producers of Hass avocado (*Persea americana* Mill.) in the central region of Veracruz State detected atypical damage associated with infestation by thrips. Larvae and adults were feeding on developing fruits, causing wounds in the epidermis, producing necrotic spots, and reducing commercial value. Insects were collected from affected fruits, mounted, and identified using specialized keys. Images of damage were obtained, and numbers of affected trees and fruits were quantified. Thrips specimens were identified as *Pseudophilothrips perseae* (Thysanoptera: Phlaeothripidae) with an incidence of 1.6% in the orchard examined. Damage by *P. perseae* in this work differed from that usually caused by other thrips species that feed and oviposit on tender tissues of avocado trees. We reported for the first time, damage caused by *P. perseae* in developing avocado fruits. More study is necessary to determine distribution and potential to increase in abundance and affect crop production.

Mexico is the main producer of avocado (*Persea americana* Mill.) worldwide, with 2.3 million tons produced annually (SIAP 2022). Avocado crops are mostly in western states of the country (Jalisco, Michoacán, and Nayarit) and in recent years spread to other states such as Veracruz where more than 1,000 ha currently are planted (SEDARPA 2022). Among the main pests that affect yield and quality of avocado are several species of phytophagous thrips that cause flowers to drop prematurely and lesions in the exocarp of newly formed fruits that could allow access to opportunistic pathogens (Hoddle et al. 2002). In Mexico, 73 thrips species associated with avocado have been recorded, of which *Scirtothrips perseae*, *Frankliniella occidentalis*, and *Neohydatothrips signifer* (Thysanoptera: Thripidae) are most important for dispersion, abundance, and damage (Bravo-Perez et al. 2018). Although avocado cultivation is recent in Veracruz State, typical damage by thrips has been recorded in inflorescences and newly formed fruits, and *Frankliniella gardeniae* and *S. perseae* were detected as potential pests of the crop (López-Lima et al. 2020). During December 2020, avocado producers in the central region of

¹Thysanoptera: Phlaeothripidae

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Veracruz detected atypical damage associated with thrips in developing fruits. Larvae and adults were feeding on fruits 5 to 10 cm in diameter, causing lesions in the exocarp as dark-brown to black necrotic spots slightly depressed (Fig. 1a-c). To identify the species and incidence of thrips causing damage, 3,000 fruits were inspected on 100 trees around the area where the damage was detected in an avocado orchard cv. 'Hass' ($19^{\circ}39'14''$ N $96^{\circ}48'05''$ W) (1,077 m above sea level) at Tepetlán municipality in central Veracruz State. A drag technique with soapy solution on a funnel attached to a 50-ml polypropylene tube was used to directly collect thrips from affected fruits (Alarcón et al. 2020). Specimens were separated from the soapy solution with a 100 μm sieve and put in 15-ml polypropylene tubes containing 70% ethanol. Subsequently, adults were dehydrated, cleared, and mounted with Canada balsam on glass slides (Bravo-Pérez et al. 2018). Specialized taxonomic keys were

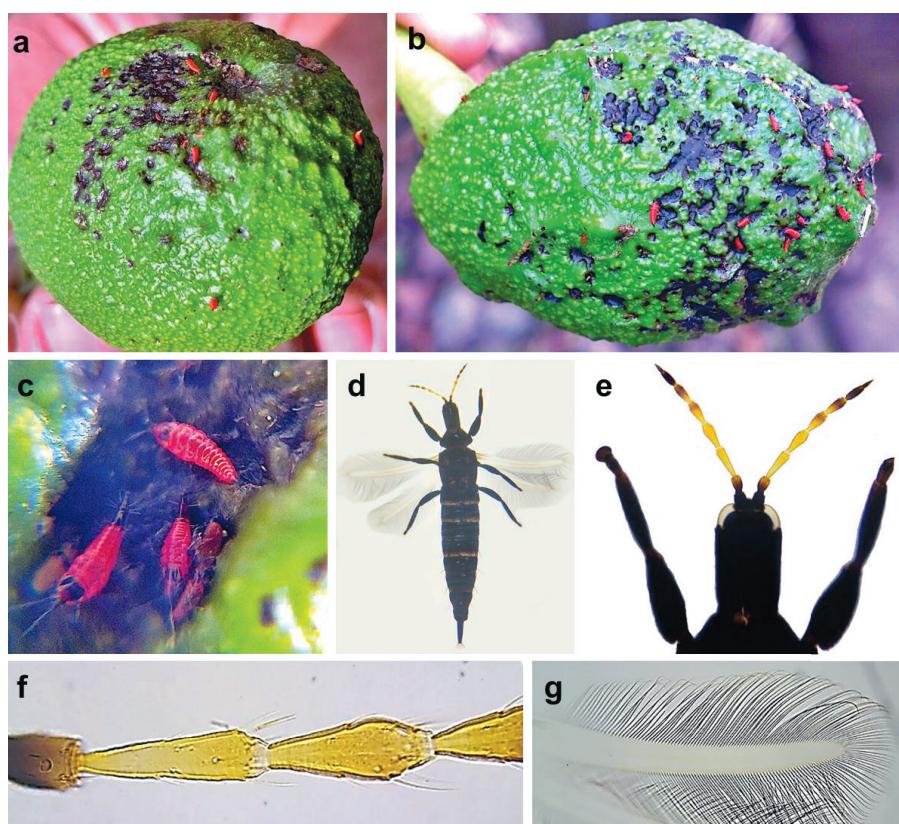


Fig. 1. *Pseudophilothrips perseae*. a-b) Damage and larvae on the avocado fruit, c) larvae aggregated in a lesion on the fruit, d) adult female 40X, e) adult showing the elongated head and antennal segments coloration 100X, f) antennal segments 3 and 4 yellow showing sense cones 400X, g) forewing showing duplicate cilia 100X.

used to identify the thrips (Mound and Kibby 1998, Mound et al. 2019). Specimens were deposited in the entomological collection of the Parasitology and Biological Control Laboratory of Faculty of Agricultural Science of the University of Veracruz (UV), Xalapa, Veracruz, Mexico.

The only thrips species collected from fruits was *Pseudophilothrips perseae* Watson (Thysanoptera: Tubulifera: Phlaeothripidae). The specimens had typical characteristics of the species such as body dark brown 4.5 to 5 mm long (Fig. 1d), with an elongated head (Fig. 1e), antennal Segment 3 yellow with a sense cone, Segment 4 slightly darker yellow with three sense cones (Fig. 1f), Segments 5-6 yellow at the base, and Segment 8 thin and slightly compressed at the base. Pronotum with five pairs of major setae with blunt apices. Metanotum with narrow and elongated reticulation. Wings well developed, pale in color with slight shading on margins, with 25 duplicated cilia (Fig. 1g).

Damage was observed in 5% of the sampled trees, with an average of 15 larvae per fruit and 10 damaged fruits per tree, which represents approximately an incidence of 1.6% in the orchard. Larvae probably have aposematic behavior to reduce predation, because they have conspicuous red color contrasting with a black terminalia and remain exposed and aggregated on the fruit surface (Hatle and Salazar 2001) (Fig. 1c).

Damage by *P. perseae* differed from those usually caused by species of Thripidae (Thysanoptera: Terebrantia) associated with avocado trees, which feed and oviposit on tender tissues such as leaf buds, inflorescences, and newly formed fruits, causing wounds that later develop into scars and ridges (Hoddle et al. 2002). Characterizing damage by *P. perseae* reduces the risk of confusion by environmental factors, fungal diseases (e.g., anthracnose caused by *Colletotrichum* spp.), or by other groups of insects (Ramírez-Gil et al. 2020).

The *Pseudophilothrips* genus has 13 species distributed on the American continent (Mound et al. 2019), with some species of agricultural and environmental importance such as *Pseudophilothrips ichini* and *Pseudophilothrips gandolfoi* (both Thysanoptera: Phlaeothripidae), potential weed biological control agents (Mound et al. 2010, Wheeler et al. 2017). *P. perseae* is native to Central America and was originally described in association with *P. americana* in Honduras and Guatemala (Hoddle et al. 2002, Mound et al. 2010). It was recently recorded as a phytophagous thrips associated with *P. americana* in Colombia, although not abundant (Cano-Calle et al. 2021). In Mexico, it was recorded in 1999 in colonies of larvae and adults on the underside of leaves of "criollo" avocado, in Nuevo León, Veracruz, and Tabasco states (Johansen et al. 1999). In general, *P. perseae*, is included as one of the thrips pests of avocado in Mexico, along with *S. perseae*, *Heliothrips haemorrhoidalis*, and *Frankliniella* spp., although there are no data to support this (Hernández et al. 2000). Its presence in Hass avocado crops was recorded only at Piste and Valladolid in Yucatan State (Hoddle et al. 2002) and in four orchards in Nayarit State as part of phytophagous thrips associated with avocado inflorescences. Although it was abundant at two sites, it is not thought to cause significant damage (Urías-López et al. 2007, Cambero et al. 2010). Thrips behavior in previous reports differed from that in this study where *P. perseae* was only on fruits and in low incidence, which classifies it as a secondary pest in Hass avocado orchards in the area. But, it causes considerable damage to fruit, reducing marketable production. More studies are necessary to determine its distribution and potential to increase in abundance and affect crop production.

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References Cited

- Alarcón-Utrera, D., D. López-Lima, D. Desgarennes, C. R. Cerdán-Cabrera, G. Alvarado-Castillo, and G. Carrión. 2020. A device for collecting thrips (Thysanoptera) from medium and tall-sized trees. Southwest. Entomol. 45: 129-136. <https://doi.org/10.3958/059.045.0114>
- Bravo-Pérez, D., M. T. Santillán-Galicia, R. M. Johansen-Naime, H. González-Hernández, O. L. Segura-León, D. L. Ochoa-Martínez, and S. Guzmán-Valencia. 2018. Species diversity of thrips (Thysanoptera) in selected avocado orchards from Mexico based on morphology and molecular data. J. Integr. Agric. 17: 2509-2517. [https://doi.org/10.1016/S2095-3119\(18\)62044-1](https://doi.org/10.1016/S2095-3119(18)62044-1)
- Cambero, O. J., R. Johansen, A. Retana, O. García, M. Cantú, y C. Carvajal. 2010. Trips (Thysanoptera) de aguacate (*Persea americana*) en Nayarit, México. Rev. Colom. Entomol. 36: 47-51.
- Cano-Calle, D., C. I. Saldamando-Benjumea, C. X. Moreno-Herrera, and R. E. Arango-Isaza. 2021. Morphological and molecular analysis of thrips (Thysanoptera: Thripidae) diversity on avocado and dandelion: new species records for Colombia and limitations for molecular differentiation of two species. Rev. Colom. Entomol. 47: e10754. <https://doi.org/10.25100/socolen.v47i2.10754>
- Hatle, J. D., and B. A. Salazar. 2001. Aposematic coloration of gregarious insects can delay predation by an ambush predator. Environ. Entomol. 30: 51-54. <https://doi.org/10.1603/0046-225X-30.1.51>
- Hernández, H. G., R. M. Johansen, L. G. Corona, A. S. Martínez-Castro, E. E. Venegas, F. D. De Anda, y A. R. Valle de la Paz. 2000. Plagas de aguacate, pp. 117-136. In D. Téliz [ed.], El Aguacate y su Manejo Integrado. Ediciones Mundiprensa, México.
- Hoddle, M. S., S. Nakahara, and P. A. Phillips. 2002. Foreign exploration for *Scirtothrips perseae* Nakahara (Thysanoptera: Thripidae) and associated natural enemies on avocado (*Persea americana* Miller). Biol. Con. 24: 251-265.
- Johansen, R. M., G. A. Mojica, y B. G. Ascención. 1999. Introducción al conocimiento de los insectos tisanópteros mexicanos, en el aguacatero (*Persea americana* Miller). Revista Chapingo Serie Horticultura 5: 279-285.
- López-Lima, D., D. Desgarennes, M. Herrera, D. Alarcón, and G. Carrión. 2020. Diversity of thrips (Thysanoptera) associated with avocado orchards in central Veracruz México. J. Entomol. Sci. 55: 141-145. <https://doi.org/10.18474/0749-8004-55.1.141>
- Mound, L. A., and G. Kibby. 1998. Thysanoptera an Identification Guide, CAB International, UK.
- Mound, L. A., G. S. Wheeler, and D. A. Williams. 2010. Resolving cryptic species with morphology and DNA; thrips as a potential biocontrol agent of Brazilian peppertree, with a new species and overview of *Pseudophilothrips* (Thysanoptera). Zootaxa 2432: 59-68.

- Mound, L. A., M. S. Hoddle, and A. Hastings. 2019. Thysanoptera California - Thrips of California. Lucidcentral.org, Identic Pty Ltd, Queensland, Australia.
- Ramírez-Gil, J. G., J. O. López, and J. A. Henao-Rojas. 2020. Causes of Hass avocado fruit rejection in preharvest, harvest, and packinghouse: economic losses and associated variables. Agronomy 10: 8. <https://doi.org/10.3390/agronomy10010008>
- SEDARPA (Secretaría de Desarrollo Agropecuario, Rural y Pesca). 2022. Anuario estadístico de la producción agrícola. <http://www.sigver-agroalimentario.sedarpa.gob.mx/ligmpa/principal/anioagricola> Accessed 17 March 2022.
- SIAP (Sistema de Información Agroalimentaria y Pesquera). 2022. Anuario Estadístico de la Producción Agrícola. <https://nube.siap.gob.mx/cierreagricola/> Accessed 19 February 2011.
- Urías-López, M. A., S. Salazar-García, and R. Johansen-Naime. 2007. Identificación y fluctuación poblacional de especies de trips (Thysanoptera) en aguacate 'Hass' en Nayarit, México. Revista Chapingo Serie Horticultura 13: 49-54
- Wheeler, G. S., V. Manrique, W. A. Overholt, F. McKay, and K. Dyer. 2017. Quarantine host range testing of *Pseudophilothrips ichini*, a potential biological control agent of Brazilian peppertree, *Schinus terebinthifolia*, in North America and Hawaii. Entomol. Exp. Appl. 162: 204-217. <https://doi.org/10.1111/eea.12506>

Diversidad de Scolytinae¹ en Huertos de Aguacate en el Estado de México**Diversity of Scolytinae¹ in Avocado Orchards in the State of Mexico**

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Resumen. Algunas especies de Scolytinae han tomado importancia en el cultivo de aguacate al ser vectores de hongos fitopatógenos. El presente estudio se realizó en tres huertos de aguacate ubicados en diferentes altitudes para conocer la diversidad y riqueza de escolitinos en Coatepec Harinas, Estado de México, especies de que pueden ser vectores de hongos fitopatógenos considerados cuarentenarios. Se recolectaron 1,714 escolitinos de 22 géneros y 49 especies en trampas tipo Lindgren cebadas con atrayentes a base de etanol, alfa-copaeno, y querciverol. De éstos, 27 son nuevos registros para el Estado de México. La prueba de índices de Shannon-Wiener (H'), Margalef (D_{Mg}), y de equidad de Pielou (J'), registraron la mayor diversidad de especies de escolitinos en Meyuca de Morelos (1890 msnm) con $H' = 2.075$, $D_{Mg} = 5.014$, y $J' = 0.5836$, y la mínima en San Martín El Salto (2470 msnm), con $H' = 0.4495$, $D_{Mg} = 2.304$, y $J' = 0.1660$. Los géneros *Xyleborus*, *Monarthrum*, *Corthylus*, y *Gnathotrichus* registraron la mayor riqueza específica, y la abundancia más alta correspondió a *Corthylus flagellifer* Blandford, *Corthylus luridus* Blandford, *Xyleborus volvulus* Fabricius, *Phloeocleptus cristatus* Wood, y *Araptus schwarzi* Blackman, individuos que representaron el 85% de captura. Riqueza y abundancia que corresponde en mayor proporción a escarabajos ambrosiales, potenciales vectores de hongos fitopatógenos, por lo que se recomienda el monitoreo frecuente y determinar la dinámica poblacional y su relación con los factores ambientales.

Abstract. Some species of Scolytinae have become important in avocado production because they can be vectors of phytopathogenic fungi. The present study was done in three avocado orchards at different altitudes to record the diversity and richness of Scolytinae at Coatepec Harinas, State of Mexico. Scolytinae that might be vectors of fungi associated with phytopathogenic fungi are considered quarantined. Scolytinae (1,714) were collected in Lindgren traps baited with attractants based on ethanol, alpha-copaeno, and querciverol, of 22 genera and 49 species, of which 27 are new for the State of Mexico. Significant differences were recorded between the sites, and the Shannon-Wiener index (H'), Margalef (D_{Mg}), and Pielou equity (J') indices tests

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recorded the greatest diversity of Scolytinae at Meyuca de Morelos (1890 masl) with $H' = 2.075$, $D_{Mg} = 5.014$, and $J' = 0.5836$, and the minimum at San Martín El Salto (2470 masl), with $H' = 0.4495$, $D_{Mg} = 2.304$, and $J' = 0.1660$. The genera *Xyleborus*, *Monarthrum*, *Corthylus*, and *Gnathotrichus* had the greatest species richness. Greatest highest abundance corresponded to *Corthylus flagellifer* Blandford, *Corthylus liridus* Blandford, *Xyleborus volvulus* Fabricius, *Phloeocleptus cristatus* Wood, and *Araptus schwarzi* Blackman, individuals that accounted for 85% of the total captures. Richness and abundance corresponded in greater proportion to ambrosial beetles, potential vectors of phytopathogenic fungi. Therefore, frequent monitoring is recommended to determine population dynamics and their relationship with environmental factors.

Introducción

Los escolitinos han tomado gran importancia en los agroecosistemas, particularmente especies de hábitos alimenticios ambrosiales, considerando que especies de Scolytinae están afectado a árboles sanos (Carrillo et al. 2012, Crane et al. 2020). Éstos se han considerado vectores de hongos fitopatógenos que causan la muerte de árboles en cuestión de semanas o meses (Ploetz et al. 2011, Mendel et al. 2012). Por ejemplo, *Xyleborus glabratus* Eichhoff tiene una asociación simbiótica con el hongo fitopatógeno *Raffaelea lauricola* (T.C. Harr, Fraedrich and Aghaye), este último responsable de la muerte de especies de plantas de la familia Laureacea, incluyendo *Persea americana* Mill (Fraedrich et al. 2008). El hongo fitopatógeno *R. lauricola* puede ser dispersado por diversas especies nativas de escarabajos ambrosiales donde se incluye a *X. volvulus*, *Xyleborus affinis* Eichhoff, *Xyleborus ferrugineus* (Fabricius), entre otros (Carrillo et al. 2014). Se ha reportado la recuperación de hongo del género *Reffaelea* en especies de escarabajos ambrosiales nativos en Michoacán (Ángel et al. 2022). Las dos especies críticas de *Euwallacea nr fornicatus* (Eichhoff) polyphagous shothole borer y kuroshio shothole borer al dispersar sus hongos simbiontes, ha causado la muerte de ramas y árboles de diversas especies vegetales en California, incluyendo la muerte parcial de ramas de aguacate (Eskalen et al. 2012, Eskalen 2022).

En México se tiene un registro de 1,041 especies de Scolytinae (Atkinson 2021). Las investigaciones realizadas en este grupo de insectos se han enfocado principalmente a determinar su impacto como descortezadores de pinos en áreas naturales. Se han desarrollado algunos estudios sobre Scolytinae asociados a agroecosistemas, como la diversidad de especies en cacao (Pérez-de la Cruz et al. 2009a,b) y en aguacate (Ángel et al. 2019, Lázaro et al. 2020). El objetivo del presente estudio consistió en determinar la diversidad y riqueza de especies nativas de Scolytinae en tres huertos de aguacate Hass ubicados en diferentes altitudes en Coatepec Harinas, Estado de México.

Materiales y Métodos

El estudio se realizó de agosto 2018 a julio del 2020 en tres huertos de aguacate Hass en Coatepec Harinas, Estado de México: 1) Meyuca de Morelos (MM) ($18.86169^\circ N$, $-099.78679^\circ W$; 1890 msnm), 2) San Martín el Salto (SMS) ($18.95715^\circ N$, $-099.79613^\circ W$, 2470 msnm), y 3) Chiltepec de Hidalgo (ChH) ($18.91885^\circ N$, $-099.83829^\circ W$; 2429 msnm). En los tres huertos se utilizaron trampas tipo Lindgren de ocho embudos, cebadas con alfa-copaeno, querciverol y etanol. En

el depósito colector de cada trampa se le adicionaron 250 ml de propilenglicol, como solución conservadora de los insectos colectados. Se colocaron 12 trampas por huerto, las cuales se colocaron en el interior del dosel a 50 cm del suelo al depósito colector. La revisión se realizó mensualmente y los insectos capturados se colocaron en frascos con alcohol al 70%.

La determinación taxonómica de los insectos capturados se realizó con las claves dicotómicas de Wood (1982) y se cotejó con especímenes de la colección de Entomología del Colegio de Postgraduados (CEAM), y se complementó con la información de Atkinson (2021) contenida en la página web: *Bark and Ambrosia Beetles* (<http://www.barkbeetles.info/about.php>). Finalmente, la corroboración de especies fue realizada por A Equihua-Martínez y T. H. Atkinson. El material se encuentra depositado en la Colección de Entomológica-CEAM. Para determinar los índices de diversidad de Shannon-Wiener (H'), índice de diversidad de Margalef (D_{Mg}) (1958), e índice de equidad de Pielou (J') (1969) se realizó el análisis estadístico con el paquete PAST 4 (Hammer et al. 2001).

Resultados y Discusión

Se obtuvieron 1,714 escolitinos distribuidos en 22 géneros y 49 especies (Cuadro 1). De éstos, 22 especies ya han sido reportadas previamente en el Estado de México (Atkinson y Equihua 1985, 1986a; Pérez et al. 2020; Atkinson 2021), y 27 especies constituyen nuevos registros para el Estado. Con estos nuevos registros, suma un total de 161 especies de Scolytinae reportados para el Estado de México, 17.37% de la biodiversidad nacional.

Los géneros *Xyleborus* Eichhoff y *Monarthrum* Kirsch, registraron la mayor riqueza con cinco especies cada uno; seguidas por los géneros *Corthylus* Erichson y *Gnathotrichus* Eichhoff con cuatro especies (Cuadro 1). Los escarabajos ambrosiales del género *Monarthrum*, se reportan en México 50 especies (Atkinson 2021). Las cinco especies colectadas son nuevos registros para el Estado de México, con un total de 11 especies del género para el estado. *Monarthrum conversum* Wood, *Monarthrum exornatum* Schedl, y *Monarthrum fimbriaticorne* Blandford tienen registros de asociación con árboles de aguacate (Ángel et al. 2019, Lázaro et al. 2020). Especies de ambos géneros son posibles vectores de hongos fitopatógenos.

Las especies *A. schwarzii* y *C. flagellifer*, registraron la mayor abundancia con 642 y 385 individuos recolectados, que representan el 60% del total. Equihua et al. (2016) indican que esta especie se encuentra asociada a la degradación e incorporación de semillas de aguacate al suelo. *C. flagellifer* se encuentra ampliamente distribuido en México, con un amplio rango de hospederos de las familias Anacardiaceae, Compositae, Lauraceae, Leguminosae, Moracea, Rocacea, y Sapindaceae (Atkinson 2021).

De las 49 especies obtenidas, 12 han sido reportadas en *P. americana*, 15 con registros previos asociados a trapeo en huertos de aguacate, y una tanto en hospedante como en trapeo (Cuadro 1). De las especies recolectadas *X. affinis*, *X. ferrugineus*, *X. volvulus*, *M. conversum*, *M. fimbriaticorne*, *M. exornatum*, y *Corthylus detrimentosus* Schedl han sido reportadas atacando árboles productivos de aguacate (Carrillo et al. 2012, Castrejón et al. 2017, Ángel et al. 2019). Las especies *X. volvulus*, *X. affinis*, y *X. ferrugineus* están reportadas en Florida como vectores laterales del hongo cuarentenario *R. lauricola* en huertos de aguacate (Carrillo et al. 2014).

Cuadro 1. Scolytinae Colectados en Trampas Cebadas con Atrayentes Químicos en Huertos de Aguacate en el Estado de México

Table 1. Scolytinae Collected in Traps Baited with Chemical Attractants in Avocado Orchards in the State of Mexico

Especie	HA	MM	SMS	Número de insectos		
				ChH	Total	%
<i>Amphicranus rameus</i> Wood 1967	Xm	0	0	2	2	0.12
<i>Araptus dentifrons</i> Wood 1974 (T)	Fl	11	0	0	11	0.64
<i>Araptus schwarzii</i> Blackman 1942 (H, T)	Es	25	404	213	642	37.46
<i>Chramesus annexetus</i> Wood 1956 (N)	Fl	0	1	0	1	0.06
<i>Chramesus pumilus</i> Chapuis 1869 (N)	Fl	2	0	1	3	0.18
<i>Cnesinus electinus</i> Wood 1967 (N, T)	Mi	37	0	0	37	2.16
<i>Cnesinus prominulus</i> Wood 1977 (N)	Mi	0	0	1	1	0.06
<i>Cnesinus setulosus</i> Blandford 1896 (N, T)	Mi	3	0	0	3	0.18
<i>Corthylus detrimentosus</i> Schedl 1940 (H)	Xm	26	4	9	39	2.28
<i>Corthylus flagellifer</i> Blandford 1904 (H)	Xm	265	5	115	385	22.46
<i>Corthylus fuscus</i> Blandford 1904	Xm	0	1	0	1	0.06
<i>Corthylus luridus</i> Blandford 1904 (N, T)	Xm	63	0	1	64	3.73
<i>Dacnophthorus clematis</i> Wood 1971 (N)	Xm	1	0	0	1	0.06
<i>Dendrocranulus cucurbitae</i> LeConte 1879 (T)	Mi	5	0	0	5	0.29
<i>Dendrocranulus gracilis</i> Wood 1982 (N)	Mi	1	0	0	1	0.06
<i>Glochinocerus gemellus</i> Blandford 1904 (H)	Xm	0	1	0	1	0.06
<i>Gnathotrichus deleoni</i> Blackman 1942	Xm	1	1	0	2	0.12
<i>Gnathotrichus nitidifrons</i> Hopkins 1905	Xm	1	1	0	2	0.12
<i>Gnathotrichus obscurus</i> Wood 1974 (N)	Xm	1	0	0	1	0.06
<i>Gnathotrichus perniciosus</i> Wood 1967 (T)	Xm	1	0	0	1	0.06
<i>Gymnochilus reitteri</i> Eichhoff 1878 (N)	Fl	0	0	1	1	0.06
<i>Hylocurus dilutus</i> Wood 1971 (N, T)	Xi	11	0	1	12	0.70
<i>Hylocurus effeminateus</i> Wood 1956 (N)	Xi	1	0	2	3	0.18
<i>Hylocurus microcornis</i> Wood 1969	Xi	11	0	0	11	0.64
<i>Hypothenemus erectus</i> LeConte 1876 (N)	FL	1	0	0	1	0.06
<i>Hypothenemus seriatus</i> Eichhoff 1872 (N, H)	Fl	1	0	0	1	0.06
<i>Ips cribicollis</i> Eichhoff 1868	Fl	0	0	1	1	0.06
<i>Micrasis amplinis</i> Wood 1971	Xi	0	0	1	1	0.06
<i>Micracis torus</i> Wood 1971 (N, T)	Xi	2	0	3	5	0.29
<i>Micracis unicornis</i> Wood 1969 (T)	Xi	0	2	13	15	0.88
<i>Monarthrum conversum</i> Wood 1974 (N, H)	Xm	5	0	0	5	0.29
<i>Monarthrum exornatum</i> Schedl 1939 (N, H)	Xm	1	0	0	1	0.06
<i>Monarthrum fimbriaticorne</i> Blandford 1905 (N, H)	Xm	0	1	1	2	0.12
<i>Monarthrum quercicola</i> Wood 1967 (N)	Xm	6	1	4	11	0.64
<i>Monarthrum scutellare</i> LeConte 1860 (N)	Xm	3	0	2	5	0.29
<i>Phloeocleptus cristatus</i> Wood 1981 (N, H)	Fl	285	3	2	290	16.92
<i>Phloeotribus destructor</i> Wood 1969	Fl	2	0	7	9	0.53
<i>Phloeotribus frontalis</i> Olivier 1795	Fl	3	0	3	6	0.35
<i>Pityophthorus obtusipennis</i> Blandford 1904 (T)	Fl	0	8	7	15	0.88
<i>Pseudopityophthorus declivis</i> Wood 1971 (N)	Fl	0	0	1	1	0.06
<i>Pseudopityophthorus durangoensis</i> Wood 1987 (N)	Fl	2	0	0	2	0.12
<i>Pityophthorus attenuatus</i> Blackman 1942 (N, T)	Fl	1	0	0	1	0.06
<i>Scolytogenes jalapae</i> Letzner 1844 (N, T)	Fl	17	0	0	17	0.99
<i>Tricolus inornatus</i> Wood 1974 (N)	Xm	0	2	1	3	0.18
<i>Xyleborus affinis</i> Eichhoff 1868 (H)	Xm	2	0	0	2	0.12
<i>Xyleborus ferrugineus</i> (F.) 1801 (H)	Xm	7	1	1	9	0.53
<i>Xyleborus intrusus</i> Blandford 1898 (T)	Xm	0	0	1	1	0.06
<i>Xyleborus palatus</i> Wood 1974 (N, T)	Xm	1	0	0	1	0.06
<i>Xyleborus volvulus</i> (F.) 1775 (H)	Xm	76	0	3	79	4.61
Subtotal		881	436	397	1,714	
Porcentaje (%)		51.40	25.44	23.16	100	
Número de especies		35	15	26	49	

(N) Nuevo registro para el Estado de México, (H) Hospedante aguacate, (T) Registro en trampas en aguacate.

HA:Habito alimenticio, Xm:Xilomicetofágia, Fl:Fleofágia, Xi:Xilofágia, Mi:Mielofágia, Es: Espermatofágia. MM: Meyuca de Morelos, SMS: San Martin el Salto, ChH: Chiltepec de Hidalgo.

Las especies de Scolytinae colectadas, presentan diferentes hábitos de alimentación, de estas, 22 (44.9%) son xilomicetófagas, seguido de fleofágia con 15 especies (30.61%), xilofágia con seis (12.24%), mielofágia con cinco (10.20%), y espermatofágia con una especie (2.04%). El hábito alimenticio de xilomicetofágia ha sido reportado en aguacate en Michoacán (Lázaro et al. 2020), en bosque mesófilo de montaña en Veracruz (Atkinson e Ibarra 2021), y en selva tropical del Sureste de México (Atkinson y Equihua 1986a). En cambio, en ecosistemas naturales en Jalisco, Atkinson y Equihua (1986b), detectaron la fleofágia como hábito de alimentación predominante y al igual que en el presente estudio, encontraron una sola especie con hábitos de espermatofágia.

La máxima diversidad y riqueza se registró en el huerto Meyuca de Morelos (1890 msnm), de conformidad con los resultados obtenidos del índice de Shannon-Wiener $H' = 2.075$, índice de diversidad de Margalef $D_{Mg} = 5.014$, e índice de equidad de Pielou $J' = 0.5836$. Sitio en el que se registró la mayor riqueza con el 74.42% de las especies registradas en el área de estudio y una abundancia de Scolytinae de 51.40%. Los huertos ubicados en San Martín el Salto (2,470 msnm) y Chiltepec de Hidalgo (2429 msnm) registraron valores inferiores en índices de diversidad en comparación con Meyuca de Morelos, siendo el primer huerto el que registro los índices más bajos, con $H' = 0.4495$, $D_{Mg} = 2.304$, y $J' = 0.166$ (Cuadro 2). Diversidad que puede asociarse, como lo menciono Wood (1982) a la disponibilidad de alimento, considerando que en estos sitios se observó en campo menor diversidad de especies vegetales.

El presente estudio contribuye al conocimiento de la diversidad de Scolytinae asociados a atrayentes químicos en Huertos de Aguacate en el Estado de México. Los índices de mayor diversidad y abundancia (H') se registraron en el huerto Meyuca de Morelos (1890 msnm) y los sitios con mayor altitud registraron la menor abundancia y diversidad específica. El 25% de las especies recolectadas tienen como hospedante al cultivo de aguacate, el 45% de las especies recolectadas corresponden a escarabajos ambrosiales, especies potenciales de ser vectores de hongos fitopatógenos, como se ha demostrado en *X. vovulus*, *X. affinis*, y *X. ferrugineus* al ser vectores del hongo cuarentenario *R. lauricola*.

Cuadro 2. Índices de Diversidad, Riqueza y Equitatividad de Scolytinae Colectados en Trampas Cebadas con Atrayentes Químicos en Huertos de Aguacate en el Estado de México

Table 2. Diversity Index, Richness and Equitativity of Scolytinae Collected in Traps Baited with Chemical Attractants in Avocado Orchards in the State of Mexico

Sitio	Insectos totales	No. especies	ID (H')	ID (D_{Mg})	Equitatividad (J')
MM	881	35	2.0750	5.014	0.5836
SMS	436	15	0.4495	2.304	0.1660
ChH	397	26	1.5090	4.178	0.4632

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Referencias Citadas

- Ángel R., M., S. Ochoa A., S. Fernández P., G. Vázquez M., A. Equihua M., A. F. Barrientos P., M. Correa A., y J. R. Saucedo C. 2019. Identificación de escarabajos ambrosiales (Coleoptera: Curculionidae) asociados a árboles de aguacate en Michoacán, México. *Folia Entomológica Mexicana* (n.s.) 5: 80-88.
- Ángel R., M., P. P. Parra, S. Ochoa A., S. Fernández P., G. Vázquez M., A. Equihua M., A. F. Barrientos P., R. C. Ploetz, J. L. Konkol, J. R. Saucedo C., and R. Gazis. 2022. First look into the ambrosia beetle-fungus symbiosis present in commercial avocado orchards in Michoacán, Mexico. *Environ. Entomol.* 51: 385-396.
- Atkinson, T. H. 2021. Bark and Ambrosia Beetles. www.barkbeetles.info Consulta Octubre 2021.
- Atkinson, T. H., y A. Equihua M. 1985. Lista comentada de los coleópteros Scolytidae y Platypodidae del Valle de México. *Folia Entomológica Mexicana* 65: 63-108.
- Atkinson, T. H., and A. Equihua M. 1986a. Biology of Scolytidae and Platypodidae (Coleoptera) of a tropical rain forest in southeastern Mexico with an annotated checklist of species. *Ann. Entomol. Soc. Am.* 79: 414-423.
- Atkinson, T. H., and A. Equihua M. 1986b. Biology of the Scolytidae and Platypodidae (Coleoptera) in a tropical deciduous forest at Chamela, Jalisco, Mexico. *Fla. Entomol.* 69: 303-310.
- Atkinson, T. H., and L. A. Ibarra J. 2021. Corrections and additions to the checklist of bark and ambrosia beetles (Coleoptera: Curculionidae: Scolytinae, Platypodinae) of mesic montane forest in Xalapa, Veracruz, Mexico. *Coleop. Bull.* 75: 227-239.
- Carrillo, D., R. E. Duncan, and J. E. Peña. 2012. Ambrosia beetles (Coleoptera: Curculionidae: Scolytinae) that breed in avocado wood in Florida. *Fla. Entomol.* 95: 573-579.
- Carrillo, D., R. E. Duncan, J. N. Ploetz, A. F. Campbell, R. C. Ploetz, and J. E. Peña. 2014. Lateral transfer of a phytopathogenic symbiont among native and exotic ambrosia beetles. *Plant Pathol.* 63: 54-62.
- Castrejón, A. J. E., R. Montesinos M., N. Acevedo R, P. Tamez G., M. A. Ayala Z., A. M. Berlanga P., y H. C. Arredondo B. 2017. Especies de *Xyleborus* (Coleoptera: Curculionidae: Scolytinae) asociados a huertos de aguacate en Colima, México. *Acta Zoo. Mex.* (n.s) 33: 146-150.
- Crane, J. H., D. Carrillo, E. A. Evans, R. Gazis, B. Schissler, F. Ballen, y J. Wasielewski. 2020. Recomendaciones para el control y mitigación de la marchitez del laurel y sus vectores, los escarabajos ambrosia, en arboledas comerciales de aguacate en Florida. Publication #HS1360S. U.F.
- Equihua, M. A., E. G. Estrada V., M. P. Chaires G., y J. A. Acuña S. 2016. Comportamiento de *Arapthus schwartzii* Blackman (Coleoptera: Curculionidae: Scolytinae) en semillas de aguacate (Hass) en diferentes estados de madurez. *Folia Entomológica Mexicana* (n.s) 2: 33-38.

- Eskalen, A. 2022. Small Beetle, Big Problem. University of California, Riverside. <https://ucanr.edu/sites/pshb/> Accessed April 2022).
- Eskalen, A., A. González, D. H. Wang, M. Twizeyimana, and J. S. Mayorquin. 2012. First report of a *Fusarium* sp. and its vector tea shot hole borer (*Euwallacea nr. fornicatus*) causing *Fusarium* dieback on avocado in California. Plant Dis. 96: 1070.
- Fraedrich, S. W., T. C. Harrington, R. J. Rabaglia, A. E. Mayfield, J. J. Hanula, J. M. Eickwort, and D. M. Miller. 2008. A fungal symbiont of the redbay ambrosia beetle causes a lethal wilt in redbay and other Lauraceae in the Southeastern United States. Plant Dis. 92: 215-224.
- Hammer, O., D. A. T. Harper, and P. D. Ryan. 2001. PAST: Paleontological statistics software package for education and data analysis. Palaeontol. Electrónica 4: 9-41.
- Lázaro, D. M. O., A. Equihua M., J. Romero N., H. González H., D. Alvarado R., J. E. Macías S., A. Castañeda V., and T. H. Atkinson. 2020. Diversity of Scolytinae (Coleoptera: Curculionidae) at three sites in the central avocado-producing region of Michoacan, Mexico. Coleop. Bull. 74: 454-462.
- Margalef, D. R. 1958. Information theory in ecology. General Systems 3: 36-71.
- Mendel, Z., A. Protasov, M. Sharon, A. Zveibil, S. Ben Yehuda, K. O'Donnell, R. Rabaglia, M. Wysoki, and S. Freeman. 2012. An Asian ambrosia beetle *Euwallacea fornicatus* and its novel symbiotic fungus *Fusarium* sp. pose a serious threat to the Israeli avocado industry. Phytoparasitica 40: 235-238.
- Pérez, S. M., A. Equihua M., J. Romero N., O. L. Segura L., H. A. Thomas, J. A. López B., and E. R. Chamé V. 2020. New distribution and host plant records of Xyleborini (Coleoptera: Curculionidae: Scolytinae) in Mexico. Coleop. Bull. 74: 860-868.
- Pérez-de la Cruz, M., A. Equihua M., J. Romero N., J. M. Valdez C., y A. De la Cruz P. 2009a. Claves para la identificación de escolítinos (Coleoptera: Curculionidae: Scolytinae) asociados al agroecosistema cacao en el sur de México. Boletín del Museo de Entomología de la Universidad del Valle 10: 14-29.
- Pérez-de la Cruz, M., A. Equihua M., J. Romero N., S. Sánchez S., E. García L., y H. Bravo M. 2009b. Escolítidos (Coleoptera: Scolytidae) asociados al agroecosistema cacao en Tabasco, México. Neotrop. Entomol. 38: 602-609.
- Pielou, E. C. 1969. An Introduction to Mathematical Ecology. Wiley-Interscience John Wiley and Sons.
- Ploetz, R. C., J. M. Pérez M., J. A. Smith, M. Hughes, T. J. Dreaden, S. A. Inch, and Y. Fu. 2011. Responses of avocado to laurel wilt, caused by *Raffaelea lauricola*. Plant Pathol. 61: 801-808.
- Wood, S. L. 1982. The Bark and Ambrosia Beetles of North and Central America (Coleoptera: Scolytidae), a Taxonomic Monograph. The Greatbasin Naturalist Memoirs 6: 1-1359.

Bioecología del Barrenador de Ramas *Neptychodes trilineatus* L.¹. en Yaca, *Artocarpus heterophyllus* Lam.

Bioecology of the Branch Borer *Neptychodes trilineatus* L.¹ in Jackfruit, *Artocarpus heterophyllus* Lam.

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Resumen. La yaca, *Artocarpus heterophyllus* Lam., es un cultivo reciente en México de producción principalmente para exportación. El barrenador de ramas, *Neptychodes trilineatus* L., es la principal plaga. Se determinaron la incidencia de larvas en ramas y severidad, distribución espacial, y fluctuación poblacional de los adultos en Nayarit, México. El adulto se alimenta de brotes tiernos y de la cáscara de frutos en desarrollo, mientras que la larva barrená las ramas. Con una incidencia media de dos larvas por kilogramo de madera seca se le encuentra durante todo el año, a excepción de diciembre. La mayor infestación ocurre en septiembre con 5.4 larvas por kilogramo de madera. El 84.7% de los árboles presentaron síntomas de daños por barrenador. Con una incidencia media de 0.92 adultos por árbol, se recolectaron en abril, mayo, julio, agosto, noviembre, enero, y febrero. En enero se detectó la mayor infestación con tres adultos por árbol. La mayor presencia ocurrió en la región de Tecuitata con 1.7 adultos por árbol, seguido de Jalcocotán con 1.1 adultos por árbol.

Abstract. Jackfruit, *Artocarpus heterophyllus* Lam., in Mexico is a recent crop for the export market. The branch borer, *Neptychodes trilineatus* L., is the main pest of the crop. The objectives were to determine the incidence of larvae in branches and severity, spatial distribution, and population fluctuation of adults at Nayarit, Mexico in 2018-2019. Adults feed on tender shoots and skin of developing fruits, while larvae bore the branches. With an average incidence of two larvae per kilogram of wood,

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they were found throughout the year, except in December. The greatest infestation occurred in September with 5.4 larvae per kilogram of wood. Eighty-five percent of the trees showed symptoms of borer damage, with a mean incidence of 0.92 adult per tree. Adults were collected in April, May, July, August, November, January, and February. In January, the largest infestation was detected with three adults per tree. The greatest presence occurred in the Tecuitata region with 1.7 adults per tree, and 1.1 at Jalcocotán.

Introducción

La yaca, *Artocarpus heterophyllus* Lam. (Moraceae), cuyo origen es el suroeste de India, es actualmente cultivado en Asia, África, y América (Brasil, México, y el Caribe) (Muniappan et al. 2012). En México es un cultivo con un crecimiento importante y su producción se destina principalmente para el mercado de exportación (Ulloa et al. 2007, Luna et al. 2013). En 2019 la superficie cultivada fue de 1,684 hectáreas, con una producción anual de 1,504 toneladas con un valor de \$200.3 millones de pesos (SIAP 2019). Esto ha convertido a la yaca en un frutal importante en algunos estados y una opción de producción en zonas tropicales de México.

Las principales limitantes para su cultivo y producción son plagas y enfermedades, con poco conocimiento sobre ellas y en general, sobre su manejo agronómico. Rodríguez et al. (2017) reportaron varios coleópteros y hemípteros como plagas potenciales de la yaca en Nayarit destacando la cochinilla rosada del hibisco, *Maconellicoccus hirsutus* Green (SENASICA 2016). Hernández et al. (2018, 2019, 2020) mencionan a *Piezogaster odiosus* Stal., *Leptopharsa* sp., *Tetranychus pacificus* McGregor, y *Neoptychodes trilineatus* L. Hernández et al. (2020) realizaron el primer reporte en México de larvas de *N. trilineatus* (Coleoptera: Cerambicidae) y observaciones preliminares en el cultivo de yaca. Estos autores consideran a *N. trilineatus* como la principal plaga de yaca en Nayarit ya que puede infestar 80% de las ramas, llegando a provocar la muerte prematura del árbol. Al carecer de mayor información en el cultivo de yaca para el control de este insecto, los técnicos y productores realizan recomendaciones y aplicaciones de insecticidas de amplio espectro como cipermetrina y clorpirifos.

Materiales y Métodos

Para evaluar la incidencia de larvas se recolectaron ramas de yaca mensualmente con síntomas de amarillamiento en etapas iniciales y ramas secas adheridas al árbol en los municipios de San Blas y Compostela, Nayarit, principal región productora de yaca en México. Huerto 1. Ejido de Jalcocotán (21.2938 latitud norte, -105.0457 longitud oeste a 576 msnm). Huerto 2. Tecuitata (21.2626 latitud norte, -105.0857 longitud oeste a 252 msnm). Huerto 3. El Llano (21.2730 latitud norte, -105.1112 longitud oeste a 18 msnm). Huerto 4. El Capomo (21.0613 latitud norte, -105.1035 longitud oeste y a 103 msnm). El clima predominante en la zona de estudio es cálido subhúmedo con lluvias en verano, precipitaciones en los meses de julio a noviembre (García 2004). Inicialmente los huertos se seleccionaron al azar. Las ramas se cortaron con tejido sano cercano al límite de la parte seca o dañada. Se seccionaron en trozos de 30 cm y se trasladaron al laboratorio de entomología agrícola del Campo Experimental Santiago Ixcuintla, del Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias para su procesamiento. Se

pesaron y contaron el total de larvas por kilo de madera muestreada de acuerdo a lo reportado por López et al. (2015). Para la identificación de larvas se observó la presencia de un pequeño grupo de espinas en la superficie ventral del último segmento abdominal, el cual separa a *N. trilineatus* de otras especies (Horton 1917). Para determinar la densidad de larvas se muestrearon 20 árboles con la misma periodicidad, observando daños en ramas con síntomas de amarillamiento, ramas secas defoliadas y con signos de barrenador como restos de excrementos, perforaciones, galerías y presencia de larvas.

Los muestreos de adultos de *N. trilineatus* se hicieron en la totalidad del árbol, considerando este como unidad de hábitat. También se hicieron colectas directas cada dos a tres semanas de febrero de 2017 a enero de 2018, se anotó comportamiento del insecto al alimentarse, ovipositar o aparearse. En cada municipio se eligieron dos huertos con yaca de entre 8 a 10 años de edad. En cada recolecta se eligieron al azar de tres a cinco árboles por sitio de muestreo con una distancia mínima entre árboles de 30 m. Se utilizó la técnica de derribo (Ruesink y Cogan 1990). En la base del dosel de cada árbol se colocó una malla circular anti áfidos de 3 m de radio y se asperjó cipermetrina (0.4 g de ingrediente activo/litro de agua), con una aspersora motorizada (STIHL® SR-420. 56.5 cm³, Potencia 2.6/3.5 KW/CV, Stihl, S.A. de C.V. Cuatlancingo, Puebla, México), aplicando 1.5 l por árbol. Después de 40-50 min, utilizando el equipo de protección adecuado, se recolectaron los insectos que cayeron sobre la malla. Adicionalmente, el árbol se sacudió para provocar la caída de insectos adheridos en el follaje o ramas. Se realizó una comparación de medias entre el promedio mensual acumulado de adultos por árbol en cada uno de los sitios.

Resultados y Discusión

Con 2.0 ± 0.4 larvas por kilogramo de madera seca de yaca, *N. trilineatus* se encuentra prácticamente durante todo el año en Nayarit, con excepción de diciembre. La mayor incidencia ocurrió en septiembre con 5.4 larvas/kg (Tabla 1). La severidad medida como porcentaje de árboles con síntomas de daños por barrenador, osciló de 68.7%, observada en marzo, hasta 93.7 % en enero, con un promedio de $84.7 \pm 1.7\%$.

En las ramas recolectadas algunas larvas permanecieron vivas durante nueve meses, la larva se alimenta y provoca daños al cultivo, lo anterior podría incrementar el potencial de *N. trilineatus* como plaga de la yaca en México. Se recolectaron 23 machos (68%) y 11 hembras (32%). *Neoptychodes trilineatus* se encuentra distribuido en toda la zona productora de yaca en Nayarit, México. Aunque con diferencias notables (Tabla 2) entre los sitios de muestreo, la distribución es amplia. Se sabe que el cultivo de yaca en Nayarit inició en el municipio de San Blas (Luna et al. 2013) lugar donde se detectó la mayor presencia del barrenador, lo anterior podría explicar mayor abundancia, y es probable que el insecto se encuentra en fase de dispersión y adaptación hacia otras zonas donde se cultiva yaca. El sitio con mayor infestación de adultos de *N. trilineatus* fue Tecuitata, con promedio anual de 1.7 adultos/árbol, ubicado a una altura de 252 msnm, con una temperatura media anual de 25.1°C y humedad relativa de 78.4%. Se recolectaron adultos de *N. trilineatus* en los meses de julio, agosto, noviembre, y enero, en este último ocurrió el mayor promedio con 1.3 adultos por árbol.

Tabla 1. Incidencia y Severidad de Larvas de *Neptychodes trilinetaus* en Yaca (*Artocarpus heterophyllus*), en Nayarit, México, 2019-2020

Table 1. Incidence and Severity of *Neptychodes trilinetaus* Larvae in Jackfruit (*Artocarpus heterophyllus*), in Nayarit, Mexico, 2019-2020

Fecha	Número de larvas	Peso de ramas (kg)*	Incidencia (larvas/kg*)	Porcentaje de árboles infestados
feb-19	27	11.9	2.3	85.0
mar-19	11	11.5	1.0	68.7
abr-19	20	13.2	1.5	83.7
may-19	40	12.4	3.2	86.2
jun-19	27	12.5	2.2	85.0
Jul-19	18	10.2	1.8	86.2
ago-19	15	9.4	1.6	88.7
sep-19	50	9.2	5.4	83.7
oct-19	6	9.3	0.6	86.2
nov-19	7	6.3	1.1	85.0
dic-19	0	6.2	0.0	83.7
ene-20	34	12.2	2.8	93.7
Media**	21.3 ± 4.3	10.35 ± 0.7	2.0 ± 0.4	84.7 ± 1.7

*kg de madera seca muestreada. **Media ± error estándar.

Tabla 2. Acumulado de Adultos de *N. trilineatus* por Árbol de *Artocarpus heterophyllus* en Nayarit, México en 2018-2019

Table 2. Accumulated Number of Adults of *N. trilineatus* per Tree of *Artocarpus heterophyllus* in Nayarit, Mexico, 2018-2019

Mes	El Capomo	El Llano	Tecuitata	Jalcocotan
Febrero	0.3	0.0	0.0	0.0
Marzo	0.3	0.0	0.0	0.0
Abril	0.3	0.0	0.0	0.3
Mayo	0.3	0.3	0.0	0.3
Junio	0.3	0.3	0.0	0.3
Julio	0.3	0.3	1.0	0.3
Agosto	0.3	0.3	2.3	0.7
Septiembre	0.3	0.7	2.3	1.7
Octubre	0.3	1.0	2.3	1.7
Noviembre	0.3	1.0	3.3	2.0
Diciembre	0.3	1.0	3.3	2.3
Enero	0.7	1.3	5.3	3.7
**Promedio	0.4 ± 0.0b	0.5 ± 0.1b	1.7 ± 0.5a	1.1 ± 0.3ab

*Promedio de los sitios muestreados. **Medias ± error estándar con distinta letra son significativamente diferentes (LSD, $P < 0.05$).

En promedio en los cuatro sitios de muestreo, se registraron cuatro picos poblacionales. En agosto y septiembre ocurrieron 0.7 insectos adultos por árbol, en noviembre y enero 0.8 insectos adultos por árbol. La emergencia de adultos en

agosto coincide con el inicio de lluvias en la zona de estudio, es probable que la población observada en los meses posteriores sea un acumulado de los insectos adultos emergidos al inicio de las lluvias, lo anterior deberá ser estudiado y corroborado en estudios posteriores. Los adultos de *N. trilineatus* se observaron en los períodos de febrero, abril a mayo y de junio a enero.

En nuestra investigación a los adultos de *N. trilineatus* se les observó alimentándose de ramas, brotes tiernos, y frutos en desarrollo. Una práctica de manejo agronómico en la yaca en Nayarit es la poda de formación y eliminación de dominancia apical al menos una vez al año y en forma continua durante la mayor parte del año se eliminan brotes de la parte interna del dosel y los frutos que no son aptos para su venta. Creemos que esta práctica frecuente podría contribuir a inducir estrés en el árbol y estimular la infestación, en consecuencia, contribuir con el incremento poblacional de *N. trilineatus*. Esta irregularidad en la oviposición y largo ciclo de vida complicarían las prácticas de manejo de esta plaga. Respecto a *B. rufomaculata* en yaca, se menciona que presenta un ciclo de vida más corto, de 170 a 290 días, tiempo durante el cual la hembra puede depositar de 20 a 25 huevos (Rajkumar et al. 2018). Las ramas de ≥ 2.54 cm de diámetro son particularmente susceptibles al ataque por esta plaga y de acuerdo a estos autores la medida más importante de manejo es mantener los árboles con buena nutrición, libres de daños físicos por las podas y plantas enfermas.

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Referencias Citadas

- García, E. 2004. Modificaciones al Sistema de Clasificación Climática de Köppen. 5ta edición. Instituto de Geografía. Cd. de México. UNAM. <http://www.publicaciones.igg.unam.mx/index.php/ig/catalog/book/83>.
- Hernández, F. L. M., H. Brailovsky, and V. Lopez M. 2018. Report of *Piezogaster odiosus* (Stal.) (Hemiptera: Coreidae) and *Leptopharsa* sp. (Hemiptera: Tingidae) as pests on *Artocarpus heterophyllus* Lam. cultivated in Nayarit, Mexico. Pan-Pac. Entomol. 94: 85-89.
- Hernández, F. L. H., E. Montalvo G., Y. Nolasco G., P. Gutiérrez M., H. González H., y J. J. Velázquez M. 2019. La araña roja en yaca: bioecología y eficacia biológica de acaricidas. Rev. Mexicana Cienc. Agric. 10: 1393-1403. <https://doi.org/10.29312/remexca.v10i6.1825>.
- Hernández, F. L. M., H. H. González H., E. Montalvo G., Y. Nolasco G., J. J. Velázquez M., and M. de L. García M. 2020. New record of larvae of *Neoptychodes trilineatus* Linnaeus, 1771 (Coleoptera: Cerambycidae) in jackfruit *Artocarpus heterophyllus* Lam. (Moraceae). Pan-Pac. Entomol. 96: 17-20.
- Horton, J. R. 1917. Three-lined fig-tree borer. J. Agric. Res. 10: 371-382. <https://naldc.nal.usda.gov/download/IND43965906/PDF>.
- López, M. V., O. R. Vargas, I. Alia T., V. H. Toledo H., A. M. Corona L., H. Delfín G., D. Guillen S., and D. Jiménez G. 2015. Xylophagous beetles (Coleoptera:

- Buprestidae and Cerambycidae) from *Ficus carica* in Morelos, Mexico. Coleopt. Bull. 69: 780-788.
- Luna, E. G., G. Alejo S., L. G. Ramírez G., y M. L. Arévalo G. 2013. La yaca (*Artocarpus heterophyllus* Lam.) un fruto de exportación. Agroproductividad 6: 65-70. <https://revista-agroproductividad.org/index.php/agroproductividad/article/view/486>
- Muniappan, R., B. M. Shepard, G. R. Carner, and P. Aun-Chuan. 2012. Arthropod Pests of Horticultural Crops in Tropical Asia. CABI, Oxfordshire.
- Rajkumar, B. M., B. Gundappa, M. M. Tripathi, and S. Rajan. 2018. Pests of jackfruit, pp. 587-602. In Omkar [ed.], Pests and their Management. Editorial Springer, India. https://link.springer.com/chapter/10.1007/978-981-10-8687-8_18
- Rodríguez, P. M., J. Cambero C., G. Luna E., A. Robles B., y K. G. Cambero N. 2017. Entomofauna asociada al cultivo de yaca (*Artocarpus heterophyllus* Lam.) en Nayarit, México. Entomología Mexicana 4: 220-223.
- Ruesink, W. G., y M. Kogan. 1990. Bases cuantitativas del manejo de plagas: muestreo y medición, pp. 389-461. In R. L. Metcalf y W. H. Luckman [eds.], Introducción al Manejo de Plagas de Insectos. Editorial Limusa, Cd. De México.
- SENASICA (Servicio Nacional de Sanidad, Inocuidad y Calidad Agroalimentaria). 2016. Cochinilla rosada del hibisco (*Maconellicoccus hirsutus* Green). SENASICA Ficha Técnica 6. https://prod.senasica.gob.mx/SIRVEF/Contenido_Publico/Fichas%20tecnicas/Ficha%20T%C3%A9cnica%20Cochinilla%20rosada.pdf.
- SIAP (Servicio de Información Agroalimentaria y Pesquera. 2019. Anuario Estadístico de la Producción Agrícola. Secretaría de Agricultura y Desarrollo Rural (SADER). <https://nube.siap.gob.mx/cierreagricola/> Consultado 31 Enero 2022.
- Ulloa, J. A., P. Rosas, J. R. Flores, B. E. Ulloa R., y H. Escalona. 2007. Comportamiento del color de bulbos del fruto de la yaca (*Artocarpus heterophyllus*) auto estabilizados en frascos de vidrio por la tecnología de obstáculos. Cienc. Tecnol. Aliment. 5: 372-378. <https://www.redalyc.org/pdf/724/72450508.pdf>.

Aspectos Biológicos y Nuevos Registros Estatales de *Cylindrocopturus biradiatus* Champion¹ en México**Biological Aspects and New State Records for *Cylindrocopturus biradiatus* Champion¹ in Mexico**

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Resumen. El picudo de la espina del nopal, *Cylindrocopturus biradiatus* Champion, es una especie de interés agrícola asociada con *Opuntia* spp., en gran parte de México. En este estudio se determinó que los adultos tienen hábitos solitarios y se alimentan de los cladodios en la primavera. Las hembras colocan un huevo en la base de las espinas y las larvas se alimentan del tejido interno que origina a la espina, e inviernan como tales. Se determinó que esta especie presenta tres estadios larvarios. Las larvas construyen una cámara de pupación debajo de la espina y la emergencia de los adultos ocurre a inicios de la primavera. Se adiciona a Guerrero, Hidalgo, y Oaxaca como nuevos sitios de distribución de *C. biradiatus*.

Abstract. The cactus spine weevil, *Cylindrocopturus biradiatus* Champion, is a species of agricultural interest associated with *Opuntia* spp. in a large part of the Mexican territory. In this study we observed that adults have solitary habits and feed on cladodes in the spring. The female lays an egg at the base of the spines and the larva feeds on internal tissue of the cladode from which a spine originates, spending the winter inside as larva. Our results showed that *C. biradiatus* has three larval stages. There is a gallery chamber under the spine built by the larva to pupate inside and emerge as an adult in early spring. Guerrero, Hidalgo, and Oaxaca are new distribution sites for *C. biradiatus*.

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Introducción

De acuerdo con el SIAP (2021), la superficie que se destina al cultivo de nopal (*Opuntia ficus-indica* Mill., Caryophyllales: Cactaceae) en México para consumo humano es de 60,702 ha, de las cuales 47,984 y 12,718 ha tienen como destino la producción de frutos (tunas) y nopalitos (cladodios jóvenes). La creciente demanda por productos a base de nopal ha contribuido con la expansión de su cultivo a nuevas regiones con características climáticas distintas a las recomendadas para su buen desarrollo, situación que somete a las plantas a estrés, haciéndolas susceptibles al ataque de insectos fitófagos (Mena-Covarrubias 2018).

Mena-Covarrubias (2018) reportó 160 especies de insectos asociados con *O. ficus-indica*. De éstas, el picudo del nopal, *Cactophagus spinolae* Champion (Coleoptera: Curculionidae); la cochinilla del nopal, *Dactylopius opuntiae* (Cockerell) Hemiptera: Dactylopiidae), y la chinche roja, *Hesperolabops nigriceps* Reuter (Hemiptera: Miridae), están catalogadas como las plagas de mayor importancia económica en México. Otras especies como *Loxomorpha flavidissimalis* (Grote) (Lepidoptera: Crambidae), y el picudo de la espina del nopal, *Cylindrocopturus biradiatus* Champion (Coleoptera: Curculionidae), también pueden alcanzar el estatus de plagas primarias en el noreste y centro de México (Vargas-Mendoza et al. 2008, Lara-Villalón et al. 2016, Bautista-Martínez et al. 2018, González-Hernández et al. 2019).

Cylindrocopturus biradiatus es una especie nativa de México (Champion 1906) asociada con nopalares de las especies *Opuntia streptacantha* Lem., *O. tomentosa* Salm-Dyck., *O. robusta* J.C. Wend., *O. huajuapensis* Bravo, *O. microdasys* (Lehm.) Pfeiff., y *O. ficus-indica* (Mann 1969, Castañeda-Vildózola et al. 2021). Los adultos se alimentan y aparean en los cladodios del nopal. Las hembras perforan la base de las areolas de los cladodios jóvenes (3 a 4 meses de edad) para introducir un huevo. Las larvas son endófitas y consumen el tejido interno de la areola. Externamente el daño se manifiesta por la presencia de secreciones de mucilago que se acumulan y cristalizan en la base de las espinas (Vargas-Mendoza et al. 2008, Castañeda-Vildózola et al. 2021). Vargas-Mendoza et al. (2008); Mena-Covarrubias (2011), Bautista-Martínez et al. (2016), y Castañeda-Vildózola et al. (2021) concluyeron que *C. biradiatus* representa una amenaza potencial para la producción comercial del nopal en el Estado de México, Querétaro, y Zacatecas.

A la fecha se desconoce información sobre su distribución, así como aspectos de su biología y evaluación de los daños que ocasiona en el cultivo del nopal, parámetros que son básicos para el diseño de estrategias de manejo. Este reporte aporta aspectos de la biología de *C. biradiatus* en condiciones de campo y la determinación de nuevos sitios de distribución en México.

Materiales y Métodos

Los estudios para conocer aspectos biológicos de *C. biradiatus* se realizaron en Toluca, Estado de México, observando una plantación de 315 plantas de nopal sin manejo fitosanitario en la Facultad de Ciencias Agrícolas de la Universidad Autónoma del Estado de México (19° 14' 35.52" Latitud norte, 99° 24' 43.2" Longitud oeste y 2,614 msnm). Las observaciones se realizaron de noviembre de 2020 a octubre de 2021. Cada semana se recolectaron 15 cladodios con síntomas de daño ocasionados por el picudo de la espina del nopal, y se hizo una disección del tejido afectado con un bisturí para la extracción, cuantificación y preservación en alcohol

al 70% de los distintos estados de desarrollo de *C. biradiatus*. En el laboratorio se midió el ancho de las cápsulas cefálicas de las larvas para determinar el número de estadios larvarios siguiendo la metodología de medición por análisis computarizado de imágenes como la propone (Hernández-Livera et al. 2005).

En marzo y abril de 2021 fueron revisados nopalitos cultivados en huertos de traspasio sin manejo fitosanitario y en nopalitos que crecen silvestres en Guerrero, Hidalgo, Morelos, Oaxaca, Puebla, y Tamaulipas. En cada lugar se buscaron plantas de nopal con daño atribuido a *C. biradiatus*. Se colectaron 15 cladodios y se removió el tejido afectado para determinar la presencia de los estados inmaduros de *C. biradiatus*. También se capturaron manualmente adultos de *C. biradiatus* sobre las plantas de nopal. Los insectos recolectados se trasladaron al laboratorio y con base a los caracteres morfológicos externos se procedió con su determinación, empleando las descripciones de Champion (1906) y Bautista-Martínez et al. (2018).

Resultados y Discusión

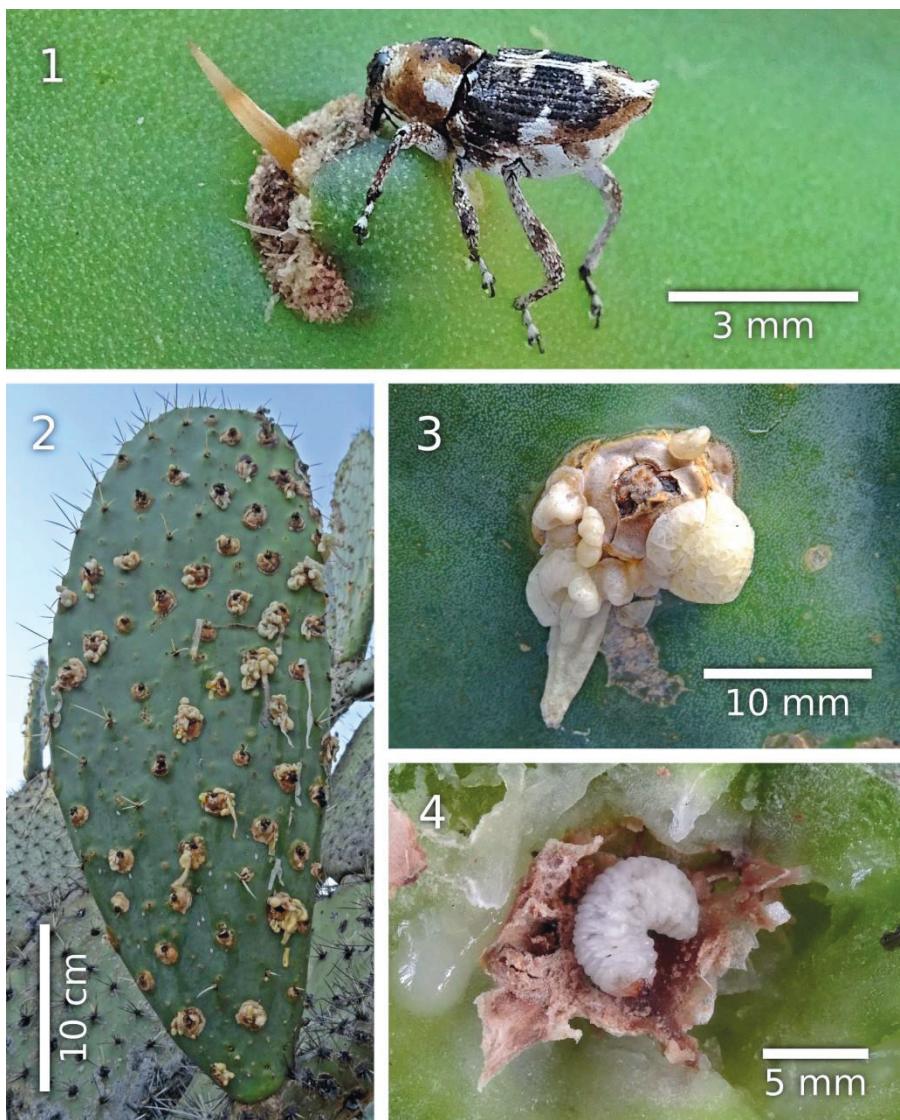
Los adultos de *C. biradiatus* son de hábitos solitarios y diurnos, fueron observados a inicios de marzo hasta mediados de julio, con la mayor incidencia en abril y mayo. En agosto no se registró presencia de adultos. Lo mismo fue reportado en Zacatecas (Mena-Covarrubias 2011) pero en Querétaro los adultos están presentes de enero hasta septiembre (Vargas-Mendoza et al. 2008).

En cada planta de nopal ($N = 55$) se encontraron de uno hasta siete picudos, distribuidos aleatoriamente. La actividad del curculiónido inicio a partir de las 09:00 hasta las 17:00 h, durante este lapso se desplazaron y alimentaron sobre los cladodios jóvenes. Después de las 17:00 h disminuyó su actividad y se ocultaron en las ranuras de las plantas de nopal. Entre abril y junio la hembra daña los cladodios, masticando el tejido de la base de la espina para perforar un orificio de oviposición (Fig. 1). Durante los días nublados o con presencia de lluvias la actividad del *C. biradiatus* fue escasa.

El promedio de espinas afectadas por cladodio fue siete, con un rango de 1 a 32 (Fig. 2). Sin embargo, los síntomas por oviposición en etapas iniciales no fueron percibidos, la manifestación del daño ocurrió a finales de septiembre, en la base de la espina se observó la secreción de una sustancia mucilaginosa que se acumuló sobre la base de la espina y con el transcurso del tiempo se cristalizó (Fig. 3), formando un grumo que representa el daño típico ocasionado por la larva de *C. biradiatus* (Fig. 4).

La distribución de frecuencias de la anchura de las capsulas cefálicas medidas a 889 larvas de distintos tamaños (321-1221 μ), sugiere que *C. biradiatus* presenta tres estadios larvarios. Nuestros resultados concuerdan con estudios realizados por Eaton 1942, Furniss 1942, Piper 1977, y Charlet 1983, que mencionan que las larvas de *Cylindrocopturus* spp., presentan tres estadios larvarios. Las larvas de primer estadio de *C. biradiatus* fueron registradas a partir de la segunda semana de agosto hasta finales de septiembre, alcanzando una longitud de 1.0 a 3.0 mm ($N = 7$), las cuales fueron difíciles de localizar por su tamaño pequeño y coloración que dificultó su ubicación en el tejido acuoso del nopal.

Las larvas de segundo estadio se presentan en octubre hasta inicios de febrero, su longitud es 4.0 a 6.0 mm ($N = 37$), su tamaño permitió una fácil localización en el tejido afectado, además su voracidad contribuyó al consumo del tejido de la areola que origina a la espina. La larva de tercer estadio alcanzó 7.0 a 9.0 mm ($N = 35$ larvas), es de complejión robusta y cabeza inmersa en el segmento

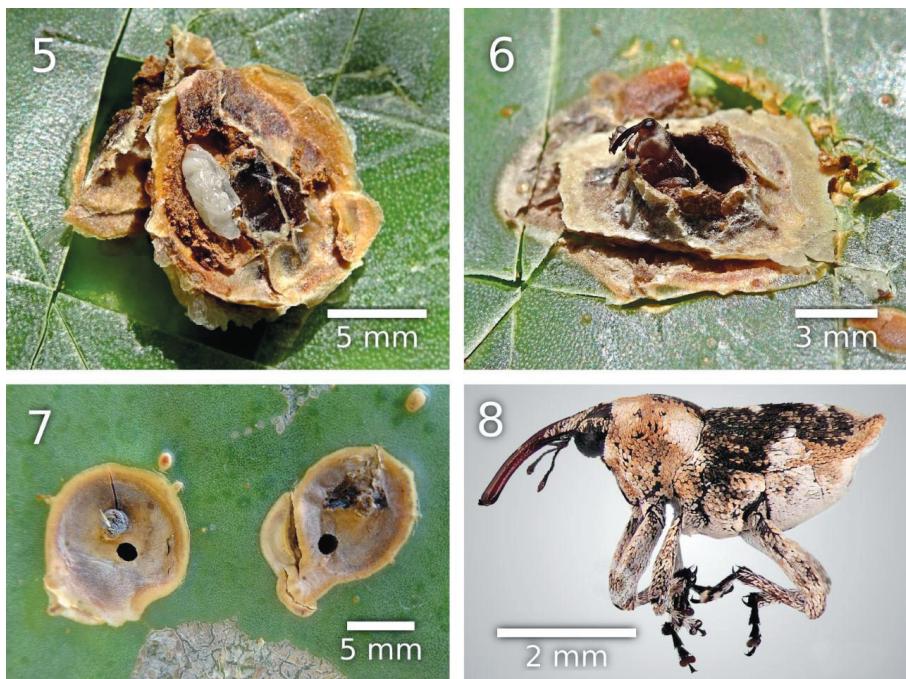


Figs. 1-4. Hembra de *Cylindrocopturus biradiatus* perforando la areola que origina la espina para ovipositar 1), cladodio infestado por *C. biradiatus* 2), daño típico ocasionado por el picudo de la espina 3), larva de *C. biradiatus* 4).

Figs. 1-4. Female of *Cylindrocopturus biradiatus* drilling to oviposit in the areola from which the spine originates 1), cladode infested by *C. biradiatus* 2), typical damage caused by the spine weevil 3), larva of *C. biradiatus* 4).

protoráxico (Fig. 5). Éstas se presentan de mediados de febrero hasta inicios de mayo, consumieron la totalidad del tejido de la areola, construyeron una cámara de pupación bajo la espina.

La presencia de pupas se registró a finales de febrero hasta mediados de mayo (Fig. 6). El tiempo de pupa a adulto es 15 días ($N = 5$). Los adultos emergieron a través de un orificio que realizaron con sus mandíbulas desde la cámara de pupación hacia el exterior (Fig. 7 y 8).



Figs. 5-8. Pupa de *Cylindrocoptus biradiatus* 5), emergencia del adulto 6), orificios de salida perforados por *C. biradiatus* 7), *C. biradiatus* en vista lateral 8).

Figs. 5-8. Pupae of *C. biradiatus* 5), emergence of adult 6), exit holes caused by *C. biradiatus* 7), *C. biradiatus* in lateral view 8).

Se capturaron 91 adultos de *C. biradiatus* sobre los cladodios de *Opuntia* spp. en cinco estados. En Janambres, Ciudad Victoria, Tamaulipas ($23^{\circ}45'41''$ N, $99^{\circ}05'37''$ W y 254 m), no se observó la presencia de adultos, ni daños atribuidos por *C. biradiatus*. La distribución actual de *C. biradiatus* está documentada en la Ciudad de México, Estado de México, Guanajuato, Michoacán, Morelos, Querétaro, Puebla, San Luis Potosí, y Zacatecas (Champion 1902, Mann 1969, Vargas-Mendoza et al. 2008, Bautista-Martínez et al. 2018, Castañeda-Vildózola et al. 2021). Este estudio adiciona a Guerrero, Hidalgo, y Oaxaca como nuevos registros, e información relevante sobre su distribución altitudinal desde los 934 hasta los 2614 msnm. Además, se adicionan nuevos registros municipales para Morelos y Puebla.

Previamente Mann (1969) documentó al picudo de la espina del nopal en el municipio de Cuernavaca, Morelos.

La información adicional asociada con los adultos de *C. biradiatus* es la siguiente: **GUERRERO**: Tepecuacuilco de Trujano, 18°10'45.7"N, 99°31'07.3"W, 934 m. Palemón, F. 17 abril 2021. 21 adultos capturados; Teloloapan, 18°14'36.2"N, 99°54'48.8"W, 1127 m. 24 abril 2021. Palemón, F. 13 adultos capturados. **HIDALGO**: Epazoyucán, 19°59'47.92" N, 99°40'40.5" W; 2419 m. Rodríguez-Ortega, A. 16 abril 2021. 24 adultos colectados; San Juan Tepa, Francisco I. Madero, 20°12'58.3" N, 99°03'58.0" W; 2018 m. Rodríguez-Ortega, A. 17 abril 2021. 6 adultos colectados. **MORELOS**: Amacuzac, 18°14'47.5" N, 97°58'45.7" W, 989 m. 10 abril 2021. Palemón, F. 4 Adultos capturado. **PUEBLA**: Tianguistengo, Acatlán de Osorio, 18°14'47.9" N, 98°01'16.4" W, 1163 m. 30 marzo 2021. Palemón, F. 14 Adultos capturados; Garzones, Acatlán de Osorio, 18°16'05.6" N, 98°01'44.5" W, 1268 m. 03 abril 2021. Palemón, F. 10 adultos capturados; Xayacatlán, 18°14'47.5" N, 97°58'46.0" W, 1261 m. 05 abril 2021. Palemón, F. 11 adultos capturados. **OAXACA**: Vista Hermosa, Huajuapan de León, 17°47'51.1" N, 97°45'23.6" W, 20 m. 17 abril 2021. Palemón, F. 6 adultos capturados.

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Referencias Citadas

- Bautista-Martínez, N., C. P. Illescas Riquelme, and R. W. Jones. 2018. *Cylindrocopturus* (Coleoptera: Curculionidae: Conoderinae) species associated with *Opuntia* (Caryophyllales: Cactaceae) species. Fla. Entomol. 99: 126-127.
- Castañeda-Vildózola, A., J. R. Sánchez-Pale, O. Franco-Mora, y J. Valdez-Carrasco. 2021. Nuevos registros de *Cylindrocopturus* y la asociación con nuevas especies de *Opuntia* en el Estado de México. Rev. Mex. Cienc. Agríc. 12: 151-156.
- Champion, G. C. 1906. Biologia Centrali-Americanana. Smithsonian Institution. Insecta, Coleoptera, Rhynchophora. Vol. 4, Part 5. Taylor and Francis [eds.], London, UK.
- Charlet, L. D. 1983. Insect stem fauna of native sunflower species in Western North Dakota. Environ. Entomol. 12: 1286-1288.
- Eaton, C. B. 1942. Biology of the weevil *Cylindrocopturus eatoni* Buchanan, injurious to Ponderosa and Jeffrey pine reproduction. J. Econ. Entomol. 35: 20-25.
- Furniss, R. L. 1942. Biology of *Cylindrocopturus furnissi* Buchanan on Douglas- fir. J. Econ. Entomol. 35: 853-859.
- Gaona-García, G., V. Vanoye-Eligio, M. Lara-Villalón, E. Ruiz-Cancino, G. Sánchez-Ramos, and M. A. Solís. 2020. First report in Mexico of *Eiphosoma dentator* (Fabricius) (Hymenoptera: Ichneumonidae) as a parasitoid of the cactus-feeding *Loxomorpha flavidissimalis* (Grote) (Lepidoptera: Crambidae). Proc. Entomol. Soc. Wash. 122: 515-518.
- García, E. 2004. Modificación al sistema de clasificación climática de Koeppen. Instituto de geografía. Universidad Nacional Autónoma de México. Número 6.

- González-Hernández, Á., V. López-Martínez, M. A. Solís, D. Guillén-Sánchez, and A. Burgos-Solorio. 2019. First report of range expansion of the cactus webworm, *Loxomorpha flavidissimalis* (Grote) (Pyraloidea: Crambidae), to central Mexico and central Texas, USA. *Fla. Entomol.* 102: 638-641.
- Hernández-Livera R. A., C. Llanderol-Cázares, L. E. Castillo-Márquez, J. Valdez-Carrasco, y R. Nieto-Hernández. 2005. Identificación de instares larvales de *Comadia redtenbacheri* Hamm. (Lepidoptera: Cossidae). *Agrociencia* 39: 539-544.
- Jones, R. W., y J. Luna-Cozar. 2007. Lista de las especies de Curculionoidea (Insecta: Coleoptera) del estado de Querétaro, México. *Acta Zool. Mex.* 23: 59-77.
- Lara-Villalón M., M. A. Solís, G. Sánchez-Ramos, and A. Mora-Olivio. 2016. The cactus webworm, *Loxomorpha flavidissimalis* (Grote, 1878) (Pyraloidea): its distribution and potential as a pest of cultivated cactus (*Opuntia ficus-indica* (L.) Mill., var. Copena V-1) (Cactaceae) in Tamaulipas, Mexico. *Proc. Entomol. Soc. Wash.* 118: 109-114.
- López-Martínez, V., N. B. Pérez-de la O, I. I. Ramírez-Bustos, I. Alia-Tejacal, and D. Jiménez-García. 2016. Current and potential distribution of the cactus weevil, *Cactophagus spinolae* (Gyllenhal) (Coleoptera: Curculionidae), in Mexico. *Coleop. Bull.* 70: 327-324.
- Mann, J. 1969. Cactus-feeding insects and mites. Smithsonian Institution, Museum of Natural History. United States National Museum Bulletin 256. Smithsonian Institution Press, Washington, DC.
- Mena-Covarrubias, J. 2011. Insectos plagas del nopal: como tomar decisiones con un enfoque de manejo integrado. *RSPyN* 12: 65-74. <https://respyn.uanl.mx/index.php/respyn>
- Mena-Covarrubias, J. 2018. Insectos plaga del nopal, pp. 129-138. In P. Inglese, C. Mondragón-Jacobo, A. Nefzaoui, y C. Sáenz [eds.], *Ecología del Cultivo, Manejo y Usos del Nopal. 1^a ed.. Organización de las Naciones Unidas para la Alimentación y la Agricultura, Centro Internacional de Investigaciones Agrícolas en Zonas Áridas*, Roma.
- Palafox-Luna, J. A., E. Rodríguez-Leyva, J. R. Lomelí-Flores, A. L. Vigueras-Guzmán, and J. M. Vanegas-Rico. 2018. Life cycle and fecundity of *Dactylopius opuntiae* (Hemiptera: Dactylopiidae) in *Opuntia ficus-indica* (Caryophyllales: Cactaceae). *Agrociencia* 52: 103-114.
- Piper, G. L. 1977. Biology and immature stages of *Cylindrocopturus quercus* (Say) (Coleoptera: Curculionidae). *Coleopt. Bull.* 31: 65-72.
- Ruiz-Machuca, M., M. Palomares-Pérez, S. Ramírez-Alarcón, E. Rodríguez-Leyva, E., y H. Brailovsky. 2010. Nuevos registros de *Hesperolabops nigriceps* Reuter (Hemiptera: Miridae) en el oriente del Estado de México. *Rev. Mex. Cienc. Agríc.* 1: 627-630.
- Salas-Araiza, M. D., Ch. W. O'Brien, and J. Romero-Nápoles. 2001. Curculionoidea (Insecta: Coleoptera) from the State of Guanajuato, Mexico. *Insecta Mundi* 15: 45-57.
- SIAP (Sistema de Información Agroalimentaria y Pesquera). 2021. Anuario Estadístico de la Producción Agrícola 2019 en México. https://nube.siap.gob.mx/cierre_pecuario/
- Soto-Hernández M., P. Reyes-Castillo, O. García Martínez, y S. Ordaz-Silva. 2016. Curculiónidos de diversas localidades de la república mexicana (Coleoptera: Curculionoidea). *Acta Zool. Mex.* 32: 62-70.

- Vargas-Mendoza, A., A. Flores-Hernández, y J. F. Basaldúa-Suárez. 2008. Dinámica poblacional de las principales plagas de nopal *Opuntia* spp. en la zona semiárida de Querétaro. Revista Chapingo Serie Zonas Áridas 7: 21-27.
- Wibmer, G. J., and Ch. W. O'Brien. 1989. Additions and corrections to annotated checklists of the weevils (Curculionidae sensu lato) of North America, Central America and the West Indies. Southwest. Entomol. 14 Suppl. 13: 1-49.

Abundance of *Diaphorina citri* Kuwayama¹ Associated with Temperature and Precipitation at Tamaulipas, Mexico**Abundancia Estacional de *Diaphorina citri* Kuwayama¹ Asociada a la Temperatura y Precipitación en el Tamaulipas, México**

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Abstract. Presence and abundance of *Diaphorina citri* Kuwayama are influenced by multiple biotic and abiotic components. We used trapping at the Regional Control Area (ARCO) to study the impact of temperature and rainfall on *D. citri*. The study during 2015-2018 was at ARCO 3 in the municipalities of Ciudad Victoria and Güémez, Tamaulipas, Mexico. *D. citri* was collected by yellow sticky traps, and temperature and precipitation were recorded every month. The previously ordered data were analyzed by multiple correspondence. Temperatures between 22 to 28°C and rainfall less than 22 mm in the study area were optimal for *D. citri*.

Resumen. En campo, la presencia y abundancia de *Diaphorina citri* es influenciada por múltiples componentes bióticos y abióticos. En el presente trabajo se determinó la participación de la temperatura y precipitación pluvial en la abundancia poblacional de *D. citri*. El trabajo fue desarrollado durante el periodo 2015-2018 en el Área Regional de Control (ARCO) número 3 localizada en los municipios de Ciudad Victoria y Güémez, Tamaulipas, México. En el ARCO, el trampío de *D. citri* se realizó con trampas pegajosas amarillas y se registró la temperatura y precipitación por mes. Los datos previamente ordenados fueron sometidos a un análisis de correspondencia múltiple. Se encontró que temperaturas entre 22 a 28°C y precipitaciones menores a 22 mm fueron las óptimas para mantener altas poblaciones de *D. citri* en la zona de estudio.

Introduction

Diaphorina citri Kuwayama (Hemiptera: Liviidae) is the main vector of Huanglongbing disease caused by *Candidatus Liberibacter* spp. (Flores-Ramírez et al. 2017). In addition to indirect damage by transmission of the pathogen, *D. citri* nymphs and adults cause direct loss by feeding on leaves, petioles, axillary buds, and tender shoots (Alemán et al. 2007).

Huanglongbing was first reported in Mexico in 2009, and reports increased because of distribution of *D. citri* (Torres-Pacheco et al. 2013). In the State of

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Tamaulipas, *D. citri* was detected for the first time in lemon trees (*Citrus limon* Burmeister) in 2003 (Coronado-Blanco and Ruiz-Cancino 2004). In 2014, *Candidatus Liberibacter asiaticus* bacterium and *D. citri* were found in Valencia orange trees (*Citrus sinensis* L.) at Tamaulipas (García-Ávila et al. 2021).

Huanglongbing affects commercial citrus plantations by killing trees, reducing fruit yield and quality, and increasing production costs (Farnsworth et al. 2014). Yield reductions range from 30 to 100% depending on the intensity of the disease because fruit is low quality (small, acidic, with low Brix index) and cannot be sold (Bassaneri et al. 2009).

Huanglongbing is in 728 municipalities in 24 states of Mexico; 391 municipalities produce citrus (SAGARPA 2018). According to the State Committee of Plant Health of Tamaulipas, the psyllid is in most municipalities of the State. In 2019, insects positive for the bacterium were reported at Abasolo, Güémez, Hidalgo, Llera de Canales, Mainero, and Victoria (CESAVETAM 2019). The State of Tamaulipas ranks second nationally in citrus production, with more than 40,000 ha cultivated (Azuara-Domínguez et al. 2021).

D. citri prefers Rutaceae plants. Reproduction depends on availability of small young shoots or freshly expanded tender leaves. Adults and nymphs need to feed on tender shoots to mature eggs and complete development (Tsai et al. 2002, Fernández and Miranda 2005, Stansly and Qureshi 2007, Qureshi and Stansly 2008). Infestation can occur if environmental conditions and host plant shoots are favorable (Hall et al. 2008, Moreno et al. 2008, García-Garduza et al. 2013, Hernández-Landa et al. 2013).

Ortega-Arenas et al. (2013) and Flores-Ramírez et al. (2017) studied Veracruz and Guerrero and found largest infestations by adult *D. citri* in citrus plantations coincided with greater outbreaks and favorable temperatures (25 to 29°C). Some authors reported more than 150 mm of monthly rainfall tended to significantly decrease abundance of *D. citri* because of washing effect: oviposition occurs in the upper shoots of host plants, thus leaving nymphs exposed to rain. Rain can also hurt adults before they reproduce (Aubert 1987, Hall et al. 2008, Ortega-Arenas et al. 2013, Flores-Ramírez et al. 2017). Yzquierdo-Álvarez et al. (2021) reported rainfall >500 mm did not adversely affect *D. citri*. Huanglongbing and *D. citri* in Mexico are by Regional Control Areas (ARCOs) in which control activities are used: epidemiological surveillance in commercial orchards and backyards, elimination of positive plants, systematic monitoring of *D. citri*, control interventions including with insecticide, and recovery of damaged areas with certified pest-free plants (SENASICA 2018, Yzquierdo-Álvarez et al. 2021). Effect of weather on abundance of *D. citri* is unknown in the citrus zone of Tamaulipas State although abundance is positively related to new shoots in trees of Valencia orange, *Citrus sinensis* (L.), and Italian lemon (*Citrus limon* (L.)) (Álvarez-Ramos et al. 2022). This work determined correlation between temperature and precipitation with abundance of *D. citri* at an ARCO at Tamaulipas, Mexico.

Materials and Methods

The study from January 2015 to December 2018 was at ARCO 3 between Ciudad Victoria and Güémez in the central region of Tamaulipas (Fig. 1). ARCO 3 has 4,077.61 ha planted with Italian lemon and Valencia orange. Yellow sticky traps 21 x 15 cm with a black grid of 2.5 x 2.5 cm (Ferommis®) were used to collect specimens. Twenty traps were put into each of 62 orchards registered in ARCO 3,

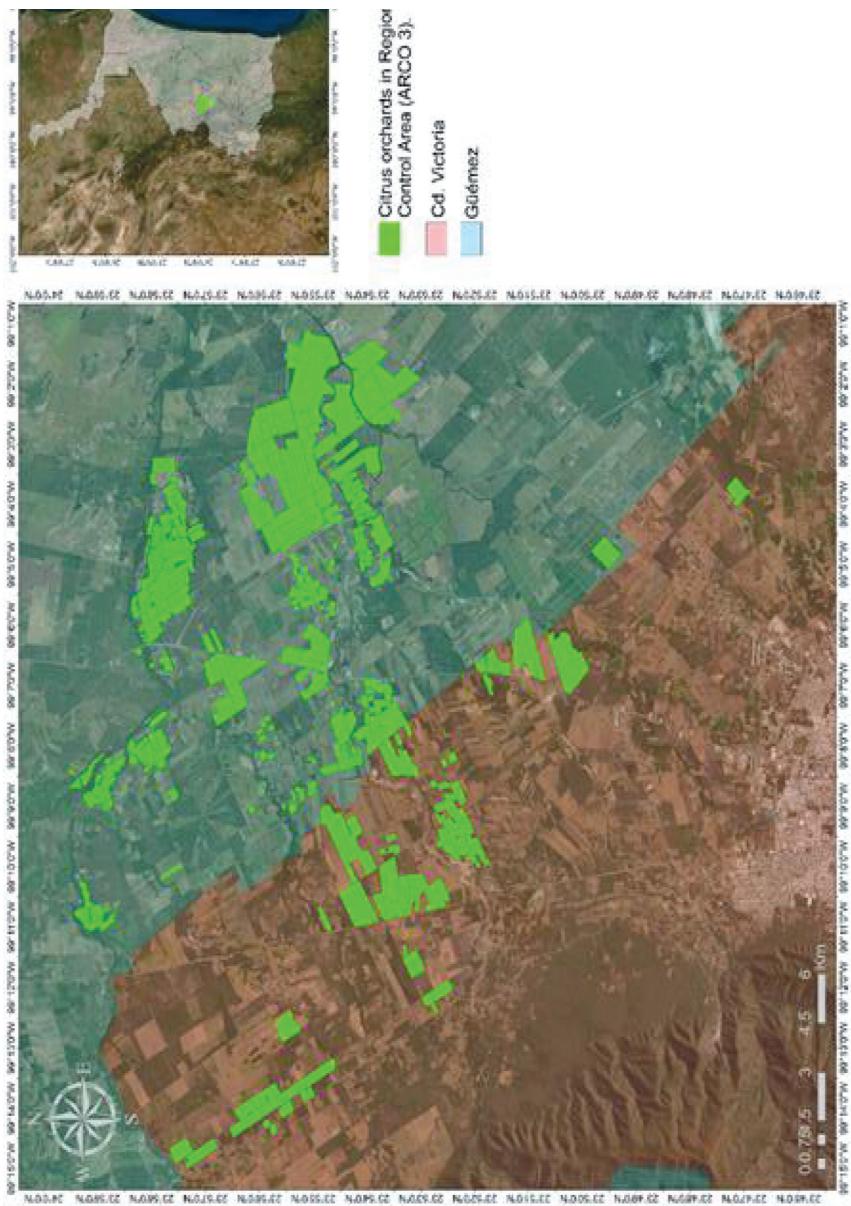


Fig. 1. Geographical location of ARCO 3 in Tamaulipas, Mexico.

for a total of 1,220 traps. The traps were oriented toward the southeastern side of the orchard alternately in the middle of the treetop in the last rows. The traps were 12 m apart and 1.2 m above the ground. Traps were collected and changed every week. The number of adult psyllids captured was counted per trap. The Diaphorina Monitoring System (SIMDIA) mobile application was used to record the number of adult psyllids captured by trap per week. Temperature and rainfall data per week were provided by the National Water Commission (CONAGUA) from weather stations closest to sampling sites.

For statistical analysis, the number of adults captured per week was ordered at intervals of 50 adults. Temperature averages were in 2°C intervals, and rainfall averages were recorded in intervals of 2 mm.

The ordered data were subjected to multiple correspondence analyses in STATISTICA version 10.2. Associations between categories were based on a two-dimensional graph with different variables reduced by increasing the distance between them (Le Roux and Rouanet 2004, Gotelli and Ellison 2013, Rodríguez-de León et al. 2017). The origin of the graph (0,0) corresponds to the average abundance of the species for each of the categories. The amount of variation in the variables to the canonical space was calculated by measuring the contribution table X^2 generated by the ACM. This metric was obtained by dividing the total (X^2) of the variable by the total (X^2) of all the variables multiplied by 100.

Results and Discussion

Multiple correspondence analysis showed significant associations between temperature and precipitation categories with adult categories captured each year (Fig. 2; $X^2 = 2063.85$, g.l = 1,024, $p = 0.000$), (Fig. 3; $X^2 = 1,807.97$, g.l = 841, $p = 0.000$), (Fig. 4; $X^2 = 2,342.36$, g.l = 1,089, $p = 0.000$), (Fig. 5; $X^2 = 1,959.42$, g.l = 784, $p = 0.000$). Association between temperature and abundance showed abundance increased when temperatures were between 22 and 28°C during the 4 years analyzed. The temperature was 24°C in 2017 when 1,350 *D. citri* adults were

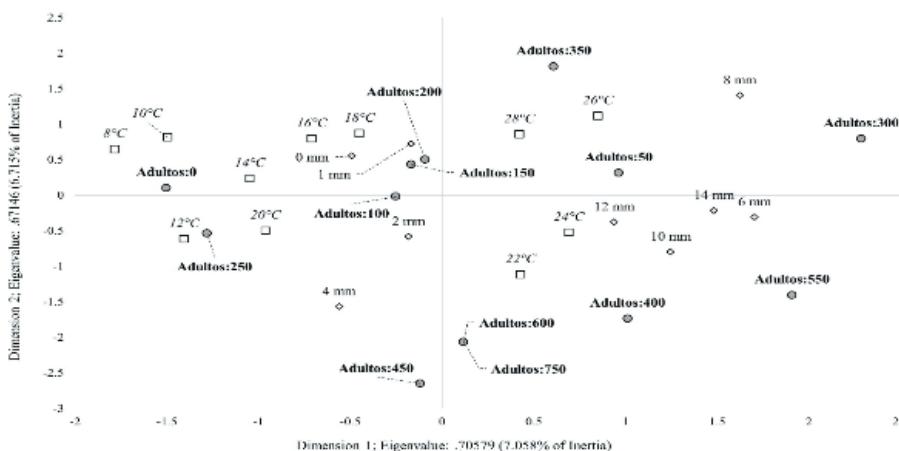


Fig. 2. Multiple correspondence analysis in the year 2015.

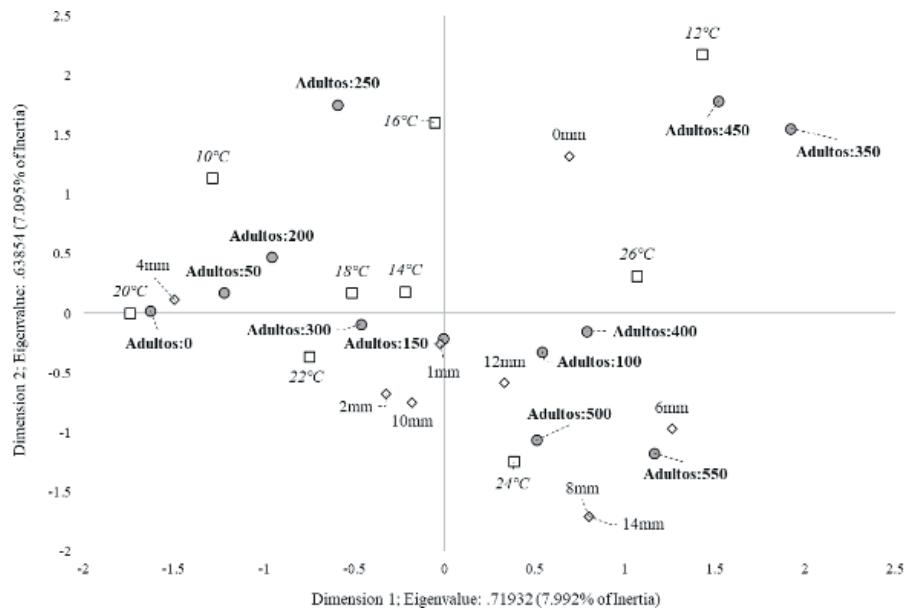


Fig. 3. Multiple correspondence analysis in the year 2016.

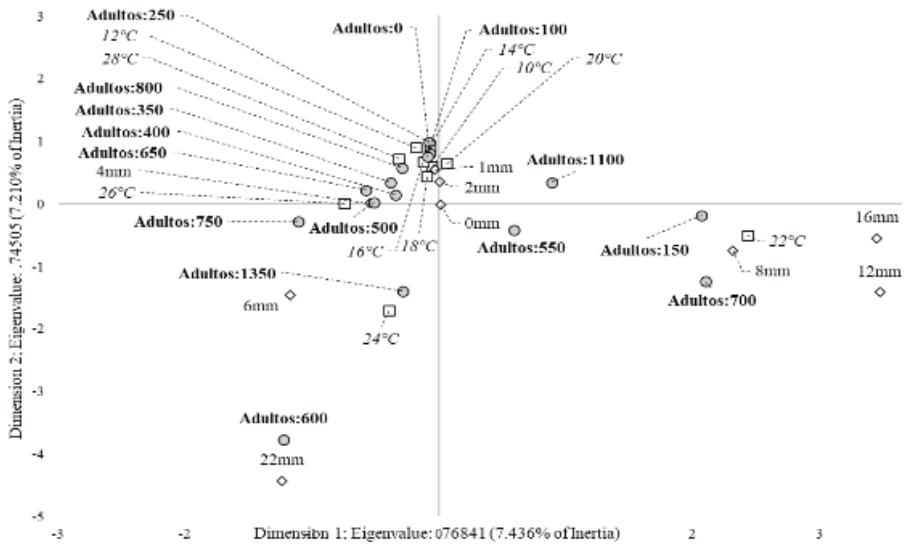


Fig. 4. Multiple correspondence analysis in the year 2017.

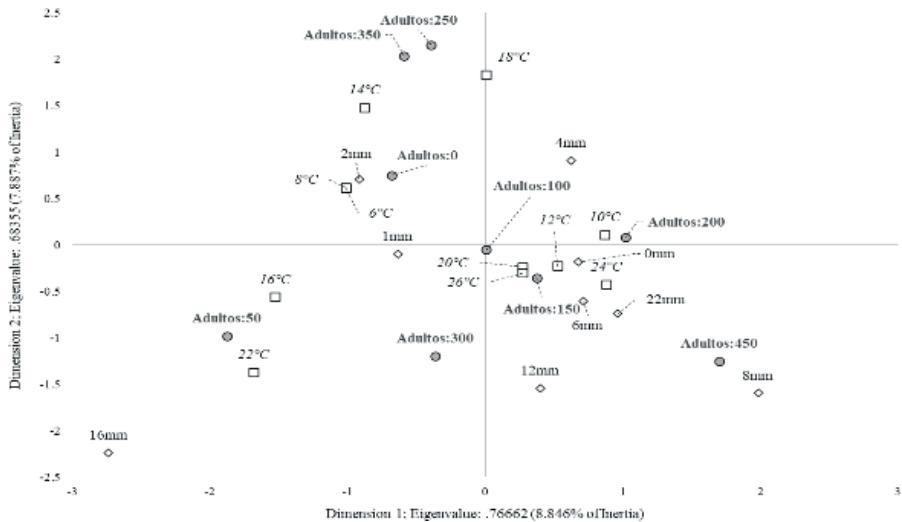


Fig. 5. Multiple correspondence analysis in the year 2018.

collected, the largest number. In 2018, greatest abundance of *D. citri* (100 to 450 adults) was associated with temperatures of 10 to 26°C (Fig. 5). In 2017, no adults were associated with temperatures between 6 and 8°C. Similarly, in 2015, temperatures between 8 and 10°C were unfavorable, with no insects detected (Fig. 2). Similar results were reported by Asplanato et al. (2011) in Uruguay who found that 12.7 to 14.7°C resulted in scarce adults of *D. citri* in the field; Alemán et al. (2007), Moreno et al. (2008), Ortega-Arenas et al. (2013), Chávez-Medina et al. (2016), Flores-Ramírez et al. (2017), and Luna-Cruz et al. (2018) reported temperatures of 22 to 30°C favored abundance of the insect. García-Garduza et al. (2013) and Chávez-Medina et al. (2016) reported abundant *D. citri* in Persian lemon (*Citrus latifolia*) plantations that coincided with abundance of shoots and warm temperatures. Liu and Tsai (2000), Nava et al. (2007), and Hodkinson (2009) also mentioned that temperature less than 10°C or warmer than 33°C prevented development of immature stages of the insect and consequently resulted in fewer adults. The authors reported optimal temperature range for *D. citri* was 25 to 28°C. Temperature data were explained by Mejía (2005) and Contreras-Servín (2014) who found warmer temperatures caused more gestation and hence more species and abundance.

Results from 2016 showed 50 to 250 *D. citri* when temperature was 10°C (Fig. 3). This confirms data reported in Florida by Halbert and Manjunath (2004) who found *D. citri* tolerated temperatures of -5°C.

Rainfall less than 14 mm led to many *D. citri* adults. The results concurred with those of Flores-Ramírez et al. (2017) who found rainfall <14 mm increased abundance of *D. citri* in the State of Guerrero. Rainfall >25 mm resulted in decreased abundance. García-Garduza et al. (2013) studied a *C. latifolia* plantation at Tabasco and reported that rainfall >50 mm negatively affected abundance of *D. citri*. Other authors found rainfall greater than 150 mm negatively affected abundance of *D. citri* in the field. The authors attributed the behavior to the knocking effect that rainfall

exerts on adults as well as washing effect that impairs nymphs and eggs deposited on the upper shoots of host plants (Aubert 1987, Hall et al. 2008, Moreno et al. 2008, Ortega-Arenas et al. 2013, Ovando and Velázquez 2013, Chávez-Medina et al. 2016, Flores-Ramírez et al. 2017). Yzquierdo-Álvarez et al. (2021) observed that *D. citri* remained abundant in the field even with rainfall >150 mm. Of note, greatest abundance of *D. citri* in 2018 (100 to 450 adults) was associated with rainfall values <22 mm. In 2017, however, 22 mm of rainfall was associated with more adults including densities of 600 (Fig. 4). Results indicated that more rainfall caused citrus plantations to produce more vegetative shoots that in turn favored insects in the field.

In citrus plantations, temperature and rainfall impact abundance of *D. citri* adults. Temperatures between 22 and 28°C and rainfall <22 mm were optimal for *D. citri* in the study area.

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References Cited

- Alemán, J., H. Baños. y J. Ravelo. 2007. *Diaphorina citri* y la enfermedad Huanglongbing: una combinación destructiva para la producción citrícola. Rev. Prot. Veg. 22: 154-165.
- Álvarez-Ramos, R., A. Azuara-Domínguez, J. H. Rodríguez-Castro, V. Zavala-Zapata, M. C. Sánchez-Borja, y J. A. Vargas-Tovar. 2022. Seasonal abundance of *Diaphorina citri* associated with the phenology of citrus crops. Rev. Mex. Cienc. Pecu. 13: 89-101.
- Asplanato, G., J. Pazos, J. Buenahora, S. Amuedo, L. Rubio, y J. Franco. 2011. El Psílido Asiático de los Cítricos, *Diaphorina citri* (Hemiptera: Psyllidae): Primeros Estudios Bioecológicos en Uruguay. INIA, Montevideo, Uruguay.
- Aubert, B. 1987. *Trioza erytreae* del Guercio and *Diaphorina citri* Kuwayama (Homoptera: Psylloidea), the two vectors of citrus greening disease: biological aspects and possible control strategies. Fruits 42: 149-162.
- Azuara-Domínguez, A., R. Álvarez-Ramos, M. O. Lázaro-Dzul, H. Vargas-Madriz, O. A. Sánchez-Flores, and S. Hernández-Contreras. 2021. First report of the whitefly *Tetraleurodes ursorum* Cockerell, 1910 (Hemiptera: Aleyrodidae) and severity of fumagin *Capnodium citri* in citrus trees in the municipality of Victoria, Tamaulipas, Mexico. Trop. Subtrop. Agroecosystems 24: 1-5.
- Bassaneri, R. B., L. H. Montesino, and E. S. Stuchi. 2009. Effects of Huanglongbing of fruit quality of sweet orange cultivars in Brazil. Eur. J. Plant Pathol. 125: 565-572.
- Chávez-Medina, J. A., G. L. Flores-Zamora, A. M. Góngora-Gómez, R. L. Gómez-Peraza, y C. B. García-Negroe. 2016. Distribución temporal de *Diaphorina citri* Kuwayama (Hemíptera: Psyllidae) en limón persa (*Citrus latifolia* Tanaka) en el municipio de Sinaloa, Sinaloa. Entomología Mexicana 3: 324-329.
- Contreras-Servín, C. 2014. Capítulo IV Condiciones climáticas asociadas al establecimiento y dispersión del Huanglongbing (HLB) y su vector la *Diaphorina citri* en México, pp. 101-117. En M. G. Galindo-Mendoza y C. Contreras-Servín [coord.], Huanglongbing y Psílido Asiático de los Cítricos: un

- Aceramiento Metodológico Multidisciplinario. Tomo 2 Colección Sanidad Vegetal. Universidad Autónoma de San Luis Potosí, México.
- Coronado-Blanco, J. M., y E. Ruiz-Cancino. 2004. Registro del "Psílido Asiático de los Cítricos", *Diaphorina citri* Kuwayama (Homóptera: Psyllidae) para México. *Folia Entomol. Mex.* 44: 165-166.
- Farnsworth, D., K. A. Grogan, A. H. C. van Bruggen, and C. B. Moss. 2014. The Potential Economic Cost and Response to Greening in Florida Citrus. *Choices*. Quarter 3. <http://choicesmagazine.org/choices-magazine/submitted-articles/the-potential-economic-cost-and-response-to-greening-in-florida-citrus>
- Fernández, M., y I. Miranda. 2005. Comportamiento de *Diaphorina citri* Kuwayama (Hemíptera: Psyllidae). Parte III: Relación entre el ciclo de vida y el brote vegetativo foliar. *Rev. Prot. Veg.* 20: 161-164.
- Flores-Ramírez, J. C., C. Aguilar-Carpio, J. A. Alcántara-Jiménez, G. Catalán-Batán, y S. Ayvar-Serna. 2017. Fluctuación poblacional del psílido *Diaphorina citri* Kuwayama (Hemíptera: Liviidae) en limón mexicano en Acapulco, Guerrero. *Acta Agr. Pec.* 3: 50-60.
- García-Ávila, C. J., F. J. Trujillo-Arriaga, A. Quezada-Salinas, I. Ruiz-Galván, D. Bravo-Pérez, J. M. Pineda-Ríos, J. G. Florencio-Anastasio, and P. L. Robles-García. 2021. Holistic area-wide approach for successfully managing citrus greening (Huanglongbing) in Mexico, pp. 33-49. En J. Hendrichs, R. Pereira, y M. J. B. Vreyzen [eds.], *Area-wide Integrated Pest Management: Development and Field Application*. CRC Press, Boca Raton, FL.
- García-Garduza, D., S. Sánchez-Soto, J. Romero-Nápoles, y J. Pérez-Flores. 2013. Fluctuación poblacional de *Diaphorina citri* (Hemiptera: Liviidae) en limón persa (*Citrus latifolia*), en Huimanguillo, Tabasco, México. *Rev. Colomb. Entomol.* 39: 201-204.
- Gotelli, N. J., and A. M. Ellison. 2013. *A Primer of Ecological Statistics*. Sinauer Associates is an Imprint of Oxford University Press, Massachusetts.
- 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. 2008. Population ecology and phenology of *Diaphorina citri* (Hemiptera: Psyllidae) in two Florida citrus groves. *Environ. Entomol.* 37: 914-924.
- Hernández-Landa, L., J. López-Collado, C. G. García-García, F. Osorio-Acosta, y M. E. Nava-Tablada. 2013. Dinámica espacio-temporal de *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) en *Murraya paniculata* (L.) Jack en Cuatláhuac, Veracruz. *Acta Zool. Mex.* 29: 334-345.
- Hodkinson, I. D. 2009. Life cycle variation and adaptation in jumping plant lice (Insecta: Hemiptera: Psylloidea): a global synthesis. *J. Nat. Hist.* 43: 65-179.
- Le Roux, B., and H. Rouanet. 2004. *Geometric Data Analysis: from Correspondence Analysis to Structured Data Analysis*. Kluwer Academic Publishers, Dordrecht, Netherlands.
- 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. Appl. Biol.* 137: 201-206.
- Luna-Cruz, A., J. J. Escamilla-García, S. I. Barreda-Pardo, and E. Loera-Alvarado. 2018. Population fluctuation of *Diaphorina citri* Kuwayana (Hemiptera: Liviidae) in the Valley of Apatzingán, Michoacán. *Acta Zool. Mex.* 34: 1-4.

- Mejía, M. 2005. Calentamiento global y la distribución de plagas. Boletín de la NAPPO (Ontario, Canada) 5-6.
- Moreno, M., E. Velásquez, R. Herrera, y M. Morales. 2008. Distribución espacial de *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) sobre lima persa (*Citrus latifolia* Tanaka). Fitosanidad 12: 33-37.
- Nava, D. E., M. L. Torres, M. D. Rodríguez, J. M. Bento, and J. R. Parra. 2007. Biology of *Diaphorina citri* (Hem., Psyllidae) on different hosts and at different temperatures. J. Appl. Entomol. 131: 709-715.
- Ortega-Arenas, L. D., A. Villegas-Monter, A. J. Ramírez-Reyes, y E. E. Mendoza-García. 2013. Abundancia estaciónal de *Diaphorina citri* (Hemiptera: Liviidae) en plantaciones de cítricos en Cazones, Veracruz, México. Acta Zool. Mex. 29: 317-333.
- Ovando, C. M. E., y M. J. J. Velázquez. 2013. Dinámica poblacional de *Diaphorina citri* en Limón Mexicano en la costa de Oaxaca, México. Simposio Internacional sobre HLB en Cítricos Acidos, Tecomán, Colima, México.
- Qureshi, J. A., and P. A. Stansly. 2008. Rate, placement, and timing of aldicarb applications to control Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae), in oranges. Pest Manag. Sci. 64: 1159-1169.
- Rodríguez-de León, I. R., J. V. Horta-Vega, C. S. Venegas-Barrera, M. Vásquez-Bolaños, y A. Correa-Sandoval. 2017. Actividad diurna de la familia Formicidae (Hymenoptera) en un gradiente de perturbación en la reserva de la biosfera "El Cielo", Tamaulipas. Entomología Mexicana 4: 455-461.
- SAGARPA. 2018. Informe de acciones operativas, mayo 2018, Campaña contra Huanglongbing de los cítricos. https://www.gob.mx/cms/uploads/attachment/file/340214/Informe_mayo_2018_HLB.pdf
- SENASICA (Servicio Nacional de Sanidad Inocuidad y Calidad Agroalimentaria). 2018. Manual operativo de la campaña contra el Huanglongbing de los cítricos. https://www.gob.mx/cms/uploads/attachment/file/262579/Manual_Operativo_contra_Huanglongbing_de_los_c_tricos.pdf
- Stansly, P. A., and J. A. Qureshi. 2007. Insecticidal control of Asian citrus psyllid through foliar applications on orange, 2006. Arthropod Manag. Tests 32: D10.
- Torres-Pacheco, I., J. L. López-Arroyo, J. A. Aguirre-Gómez, R. G. Guevara-González, R. Yáñez-López, M. I. Hernández-Zul, and J. A. Quijano-Carranza. 2013. Potential distribution in Mexico of *Diaphorina citri* (Hemiptera: Psyllidae) vector of Huanglongbing pathogen. Fla. Entomol. 96: 36-47.
- 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.
- Yzquierdo-Alvarez, M. E., J. A. Rincón-Ramírez, E. Loeza-Kuk, J. F. López-Olguín, L. A. Aceves-Navarro, y C. F. Ortiz-García. 2021. Fluctuación espacio-temporal de *Diaphorina citri* Kuwayama (Hemiptera: Liviidae) en limón persa (*Citrus latifolia*) en la zona citrícola de Huimanguillo, Tabasco. Acta Zool. Mex. 37: 1-14.

Depredación de *Exochomus bicolor*¹ sobre el Psílido Asiático de los Cítricos² y la Escama Verde³ bajo Condiciones de Laboratorio**Predation of *Exochomus bicolor*¹ on Asian Citrus Psyllid² and Coffee Green Scale³ under Laboratory Conditions**

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Resumen. Se evaluó la capacidad del adulto de *Exochomus bicolor* Fernández-García y Milán-Vargas como depredador de *Diaphorina citri* Kuwayama y *Coccus viridis* Green. *Exochomus bicolor* consume todos los estadios ninfales de *D. citri*. El tiempo letal medio (TL_{50}) (tiempo durante el cual fue consumido el 50% de los individuos) sobre *D. citri* fue de 2.86 h y el TL_{90} de 8.23 h. Para *C. viridis*, el TL_{50} fue de 2.01 h y el TL_{90} de 6.91 h. Los resultados obtenidos en este estudio demuestran que *E. bicolor* posee características que favorecen su función como enemigo natural de *D. citri* y *C. viridis*.

Abstract. Adults of *Exochomus bicolor* Fernández-García and Milán-Vargas can prey on *Diaphorina citri* Kuwayama and *Coccus viridis* Green. *Exochomus bicolor* consumed all the nymphal stages of *D. citri*. The mean lethal time (LT_{50}) (time during which 50% of individuals were consumed) for *D. citri* was 2.86 hours and the LT_{90} was 8.23 hours, and for *C. viridis*, the LT_{50} was 2.01 hours and the LT_{90} was 6.91 hours. These results show that *E. bicolor* can function as a biological control agent in the management of *D. citri* and *C. viridis*.

Introducción

Diaphorina citri Kuwayama (Hemiptera: Liviidae) es una de las plagas más importantes de los cítricos ya que es vector de las bacterias *Candidatus Liberibacter* spp. causantes de la enfermedad Huanglongbing (HLB), la cual ha afectado la producción citrícola a nivel mundial (Urbaneja-Bernat et al. 2020). *Diaphorina citri* es una especie nativa de Asia tropical y subtropical que se ha diseminado al continente americano, incluyendo Estados Unidos y países de Centroamérica, Suramérica, y el Caribe (Oke et al. 2020, Antolinez et al. 2021). La presencia de *D. citri* en Cuba se

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reportó desde 1999 (González-Fernández et al. 2010) y en 2006 se confirmó la presencia del HLB en la Ciudad de La Habana (Luis-Pantoja y Paredes-Tomás 2018) ocasionado un fuerte impacto. A finales de la década de los noventa, la producción citrícola de cuba ascendía a 220,000 t (Anaya Cruz 2015) para el 2017 la producción se limitó a 60,206 t (Luis-Pantoja y Paredes-Tomás 2018).

Coccus viridis Green (Hemiptera: Coccoidea) afecta a una serie de cultivos comerciales, entre ellos aguacate, apio, lima, café y naranja, lo que ocasiona un impacto negativo en el comercio (Randhawa 2016). Además, la melaza secretada por *C. viridis* en la planta hospedante favorece el crecimiento de hongos saprófitos, lo cual reduce la comerciabilidad de la fruta (Elmer et al. 1975). Grandes poblaciones de *C. viridis* pueden causar amarillamiento, defoliación, reducción de frutos y vigor de la planta (Randhawa 2016, Quek et al. 2017), y su presencia en hospedantes en viveros comerciales puede ocasionar cuarentenas del material vegetal infestado (Randhawa 2016).

Diversos depredadores de *D. citri* y de *C. viridis* pertenecen a la familia Coccinellidae. En Cuba, González-Fernández et al. (2010) registraron a los coccinélidos depredadores de *D. citri*: *Cycloneda sanguinea* (L.), *Chilocorus cacti* (L.), *Arawana cubensis* Dimmock, y *Scymnus distinctus* Casey. En Chiapas, México, se ha registrado a *Diomus lupusapudoves* Vandenberg, Iverson y Liere depredando a *C. viridis* (Vandenberg et al. 2018). En Brasil a *Azya luteipes* Mulsant depredando a *C. viridis* (Nais y Busoli 2012). El objetivo del presente estudio es evaluar al coccinélido *Exochomus bicolor* Fernández-García y Milán-Vargas como depredador de *D. citri* sobre plantas de *Murraya paniculata* (L.) Jack y de *C. viridis* en plantas de café (*Coffea arabica* L.).

Materiales y Métodos

La recolecta de los coccinélidos se llevó a cabo en el municipio Playa, La Habana, Cuba (23° 07' 02" N, 82° 25' 21" O; 23° 06' 56" N, 82° 25' 22" O; 23° 07' 05" N, 82° 24' 50" O y 23° 07' 08" N, 82° 24' 59" O) durante agosto y septiembre de 2021. Los muestreos se realizaron en jardines y patios con presencia de *M. paniculata* y *C. arabica*; donde se recolectaron adultos alimentándose de *D. citri* en plantas de *M. paniculata*, y de *C. viridis* en plantas de *C. arabica*. La identificación de los coccinélidos (Fernández-García y Milán-Vargas 2010) concluyó que todos los coccinélidos fueron *E. bicolor*.

Para determinar la capacidad depredadora de *E. bicolor* sobre *D. citri* se usó una caja de Petri de 5 cm de diámetro en la que se le colocó un disco de papel filtro, una hoja de *Citrus aurantifolia* (Christm) Swingle "limón mexicano", con un algodón humedecido en el pecíolo para mantener la turgencia del tejido vegetal, y se infectó con 10 ninfas de cada uno de los cinco instares de *D. citri* (50 individuos por caja). Se colocó una hembra adulta de *E. bicolor*, con 24 h de ayuno. Se contabilizó el número de *D. citri* depredados a las 24 h. Se realizaron cinco repeticiones del experimento.

Para determinar la capacidad depredadora sobre *C. viridis* se implementó la misma metodología que para *D. citri*, con las diferencias de que se colocó una hoja de café con un algodón humedecido en el pecíolo para mantener la turgencia del tejido vegetal y se infectó con 20 hembras adultas del cóccido. Se realizaron cinco repeticiones del experimento.

Los datos se examinaron con un análisis de varianza y las medias se compararon mediante la prueba de Tukey con nivel de confianza 95%. Se empleó el paquete estadístico InfoStat versión 2020 (Di Rienzo et al. 2020).

Con el objetivo de determinar el tiempo letal medio de *E. bicolor* sobre *D. citri*, se usó una caja de Petri de 5 cm de diámetro en la que se le colocó un disco de papel filtro, se colocaron 30 ninfas de *D. citri* (seis individuos de cada instar) y una hembra adulta de *E. bicolor* con 24 h de ayuno. Se realizaron cinco repeticiones del experimento.

Para evaluar el tiempo letal medio de *E. bicolor* sobre *C. viridis*, se tomó una caja de Petri con un disco de papel filtro y se colocaron siete ninfas de cada uno de los tres estadios ninfales, nueve hembras adultas de *C. viridis* y una hembra de *E. bicolor* con 24 h de ayuno. Se realizaron cinco repeticiones del experimento. Para cada experimento se realizó el conteo de los insectos depredados por cada hora, hasta que todos los individuos fueron consumidos.

Se determinó el tiempo durante el cual fue consumido el 50% (TL_{50}) y el 90% (TL_{90}) de la población. Para este cálculo se utilizó el modelo de probabilidad no lineal con especificación de los modelos de elección discreta (Probit). Los análisis se desarrollaron mediante el paquete estadístico InfoStat versión 2020 (Di Rienzo et al. 2020). Todos los bioensayos se realizaron durante el mes de agosto del año 2021, bajo condiciones controladas de temperatura entre 27-30°C, con 78% de humedad relativa y un fotoperiodo de 16 h luz y ocho de oscuridad.

Resultados y Discusión

Al cabo de 24 h todas las ninfas de *D. citri* fueron depredadas. Este resultado indica que *E. bicolor* es capaz de consumir indistintamente todos los estadios ninfales de *D. citri* sin diferencia significativa en el consumo promedio ($F = 0.17$, $p = 0.9518$) (Cuadro 1). En el caso de *C. viridis*, a las 24 h *E. bicolor* consumió el 83% de hembras adultas del cóccido ($p < 0.0001$).

En ambas poblaciones 100% de depredación se alcanzó a las 8 h. Sin embargo, antes de las 6 h, el depredador consumió una mayor cantidad de individuos de *C. viridis* que de *D. citri* (Fig. 1).

El adulto de *E. bicolor* no tuvo preferencia por ninguno de los instares. Se observó a *E. bicolor* consumir parcial o totalmente al quinto instar de las ninfas de *D. citri*, a diferencia de los cuatro primeros instares que fueron consumidos en su totalidad (Fig. 2A,B). Este es un aspecto importante ya que al impedir que *D. citri* llegue a convertirse en adulto, limita la dispersión del mismo. En el caso de *C. viridis*

Cuadro 1. Consumo Promedio de *E. bicolor* Sobre los Cinco Estadios Ninfales de *D. citri* Bajo Condiciones de Laboratorio

Table 1. Average Consumption of *E. bicolor* on the Five Nymphal Stages of *D. citri* Under Laboratory Conditions at Room Temperature

Estadios de ninfas <i>D. citri</i>	Ninfas consumidas (Medias ± D.E.)
N1	9.20 ± 1.10
N2	9.20 ± 0.84
N3	9.00 ± 1.22
N4	9.20 ± 0.84
N5	8.80 ± 0.84

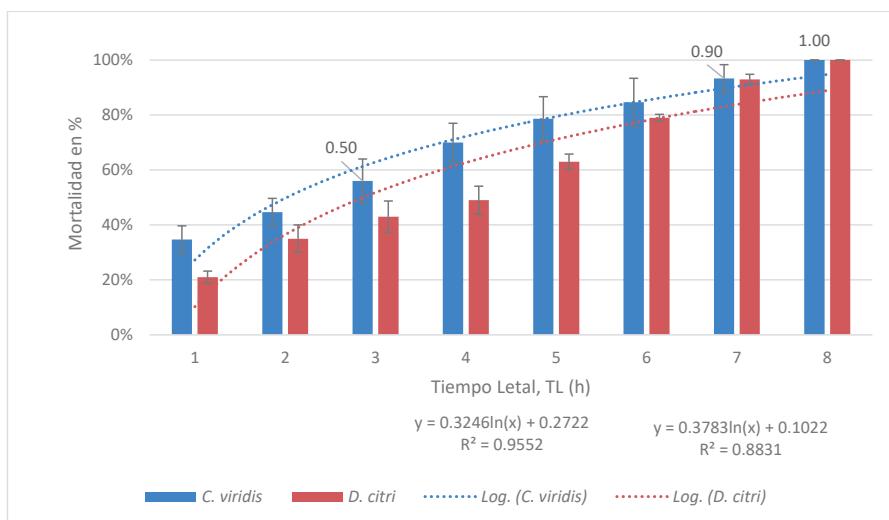


Fig. 1. Mortalidad de estadios de *Coccus viridis* y *Diaphorina citri* ocasionada por el adulto de *Exochomus bicolor*.

Fig. 1. Stage mortality of *Coccus viridis* and *Diaphorina citri* caused by adult *Exochomus bicolor*.

se observó a *E. bicolor* consumir totalmente los tres estados ninfales, mientras que a algunos adultos lo mutilaba y se alimentaba solo de la parte superior (Fig. 2C,D) y otros los consumía en su totalidad.

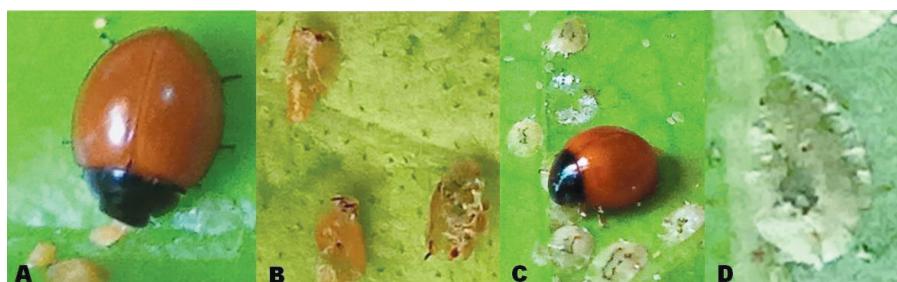


Fig. 2. A) Adulto de *Exochomus bicolor* alimentándose de ninfas de *Diaphorina citri*; B) ninfas de *D. citri* consumidas parcialmente; C) adulto de *E. bicolor* alimentándose de *Coccus viridis*; D) hembra de *C. viridis* mutilada.

Fig. 2. A) *Exochomus bicolor* adult feeding on *Diaphorina citri* nymphs; B) *D. citri* nymphs partially consumed; C) *E. bicolor* adult feeding on *Coccus viridis*; D) mutilated *C. viridis* female.

Exochomus bicolor pudo consumir parcial o totalmente el quinto instar de *D. citri*, debido al tamaño ligeramente superior de *E. bicolor* (3.0 – 3.9 mm de longitud) (Fernández-García y Milán-Vargas 2010) con respecto a *E. marginipennis* (2.5-3.6 mm de longitud) (Gordon 1985) el cual consume en menor cantidad y solo parcialmente el quinto instar (Palomares-Pérez et al. 2016). Los resultados de las observaciones realizadas en este trabajo indican que *E. bicolor* se puede alimentar de todos los estadios ninfales de *D. citri* y *C. viridis*.

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Referencias Citadas

- Anaya Cruz, B. 2015. Las cadenas productivas con impacto económico y social: el caso de los cítricos en Cuba. Econ. Desarro 154: 105-117.
- Antolinez, C. A., T. Moynear, X. Martini, and M. J. Rivera. 2021. Temperatures decrease the flight capacity of *Diaphorina citri* Kuwayama (Hemiptera: Liviidae). Insects 12: 2-11.
- Di Rienzo, J. A., F. Casanoves, M. G. Balzarini, L. González, M. Tablada, y C. W. Robledo. 2020. GrupoInfoStat, FCA, Universidad Nacional de Córdoba, Argentina. <http://www.infostat.com.ar>
- Elmer, H. S., O. L. Brawner, and W. H. Ewart. 1975. Control of brown soft scale in Central Valley. Citograph 60: 402-403.
- Fernández-García, I., y O. Milán-Vargas. 2010. Nueva especie de *Exochomus Redtenberg* (Coleoptera: Coccinellidae, Chilocorinae) de Cuba. Bol S.E.A. 47: 171-174. http://sea-entomologia.org/Publicaciones/PDF/BOLN_47/171174_BSEA47COMPLETO-3.pdf
- González-Fernández, C., M. Gómez-Pacheco, D. Hernández-Espinosa, y J. L. Rodríguez-Tapia. 2010. Entomófagos asociados a las plagas citrícolas, *Lepidosaphes gloverii* Packard (Hemiptera: Diaspididae), *Phylloconistis citrella* Stainton (Lepidoptera: Gracillariidae) y *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) en naranjo Valencia. Ctro. Agr. 37: 75-81. http://cagricola.uclv.edu.cu/descargas/pdf/V37-Numero_4/cag144101764.pdf
- Gordon, R. D. 1985. The Coccinellidae (Coleoptera) of America North of Mexico. J. N.Y. Entomol. Soc. 93: 1-912.
- Luis-Pantoja, M., y C. Paredes Tomás. 2018. La enfermedad Huanglongbing de los cítricos en Cuba. Tropicsafe <https://www.tropicsafe.eu/es/huanglongbing-disease-citrus-cuba/>
- Nais, J., and A. C. Busoli. 2012. Morphological, behavioral and biological aspects of *Azya luteipes* Mulsant fed on *Coccus viridis* (Green). Sci Agric. 69: 81-83.
- Oke, A. O., A. A. Oladigbolu, M. Kunta, O. J. Alabi, and M. Sétamou. 2020. First report of the occurrence of Asian citrus psyllid *Diaphorina citri* (Hemiptera: Liviidae), an invasive species in Nigeria. West Africa. Sci. Rep. 10: 1-8.
- Palomares-Pérez, M., B. Rodríguez-Vélez, M. A. Ayala-Zermeño, J. D. J. de la Cruz-Llanas, A. M. Mendoza-Castañeda, J. A. Sánchez-González, y E. G. Cordoba-Urtiz. 2016. Aspectos biológicos y capacidad de depredación de *Exochomus marginipennis* (LeConte) (Coleoptera: Coccinellidae) sobre *Diaphorina citri* Kuwayama (Hemiptera: Liviidae). Chil. J. Agric. Anim. Sci. 32: 102-109.

- Quek, S. P., S. Ueda, P. J. Gullan, T. Kondo, M. Hattori, T. Itioka, K. Murase, and T. Itino. 2017. Nuclear DNA based species delineations of *Coccus* scale insects in symbiosis with plants and ants, and the role of plant epicuticular wax in structuring associations. Biol. J. Linn. Soc. Lond. 120: 818-835.
- Randhawa, R. 2016. California Pest Rating for *Coccus viridis* (Green Scale) Hemiptera: Coccidae. Hemiptera, Insects, Mites & Earthworms. <https://blogs.cdfa.ca.gov/Section3162/?p=2779>
- Urbaneja-Bernat, P., D. Carrillo, and J. A. Jaques. 2020. Behavior of *Diaphorina citri*: an investigation of the potential risk to the most commonly used citrus rootstock in Europe. Entomol. Gen. 40: 79-86.
- Vandenberg, N. J., A. Iverson, and H. Liere. 2018. A new species of myrmecophilous lady beetle in the genus *Diomus* (Coleoptera: Coccinellidae: Diomini) from Chiapas, Mexico that feeds on green coffee scale, *Coccus viridis* (Green) (Hemiptera: Coccidae). Zootaxa 4420: 113-122.

Insecticidas Efectivos para el Control de Ninfas del Psílido Asiático de los Cítricos¹ en Limón Persa**Effective Insecticides for the Control of Asian Citrus Psyllid¹ Nymphs in Persian Lime**

Miguel Ángel López², Edgardo Cortez-Mondaca^{3*}, y José Isabel López-Arroyo⁴

Resumen. Se evaluaron insecticidas sintéticos y naturales con alta efectividad biológica y con reducido impacto sobre insectos no blanco, para el control del psílido asiático de los cítricos (PAC), *Diaphorina citri* Kuwayama (Hemiptera: Liviidae), en una huerta de limón Persa. Spirotetramat (75 g AI/ha), fenpiroximate (25.5 g AI/ha), y clorantraniliprol (20 g AI/ha) mostraron una mortalidad de 83.9, 85.1, y 86.6% sobre las ninfas del PAC. Ciantraniliprol a dosis de 30 y 40 g AI/ha produjo 100% de mortalidad. Flupiradifurone a 75, 112.5, y 150 g AI/ha mostró mortalidad de 91.2, 96.6, y 100%. Buprofezin 668.9 y 891.9 g AI/ha provocó mortalidad de 92.3 y 100%. Tolfenpirad a dosis de 79.5, 119.3, y 159 g AI/ha provocó mortalidad de 93.1, 100, y 100%. Los resultados obtenidos amplían el abanico de alternativas insecticidas para el manejo preventivo de la resistencia del psílido asiático de los cítricos, algunos de ellos con reducido impacto sobre organismos no blanco.

Abstract. Synthetic and natural insecticides with high biological effectiveness and reduced impact on non-target insects were evaluated for the control of the Asian citrus psyllid (ACP), *Diaphorina citri* Kuwayama (Hemiptera: Liviidae), in a Persian lime orchard. Spirotetramat (75 g AI/ha), fenpyroximate (25.5 g AI/ha), and chlorantraniliprole (20 g AI/ha) showed mortality of 83.9, 85.1, and 86.6% on ACP nymphs. While ciantraniliprole at doses of 30 and 40 g AI/ha produced 100% mortality. Flupyradifurone at doses of 75, 112.5, and 150 g AI/ha showed mortality of 91.2, 96.6, and 100% mortality. Buprofezin 668.9 and 891.9 g AI/ha caused mortality of 92.3 and 100%. Tolfenpyrad at doses of 79.5, 119.3, and 159 g AI/ha caused mortality of 93.1, 100, and 100%. These results expand the range of insecticide alternatives for the preventive management of the resistance of the Asian citrus psyllid, some of them having a reduced impact on non-target organisms.

¹*Diaphorina citri* Kuwayama (Hemiptera: Liviidae).

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Introducción

El Huanglongbing (HLB) de los cítricos, es la enfermedad considerada como la más devastadora que afecta a este tipo de árboles (Chen et al. 2009). En México, se asocia al patógeno *Candidatus Liberibacter asiaticus* (CLas), el cual fue detectado por primera vez en 2009 en Yucatán, en muestras de su principal insecto vector, el psílido asiático de los cítricos (PAC) *Diaphorina citri* Kuwayama (Hemiptera: Liviidae) (SENASICA 2009). La estrategia de manejo, está basada en la aspersión estratégica de insecticidas dos o tres veces al año, protegiendo aproximadamente 30% de la citricultura nacional. El propósito es reducir al máximo las poblaciones del PAC para disminuir y retrasar la presencia y diseminación del HLB (Cortez et al. 2011, Boina y Bloomquist 2015). Debido a que el control del PAC se basa en el control químico (Della et al. 2018, Iqbal et al. 2020), es sumamente importante utilizar insecticidas con probada efectividad, mediante evaluaciones que se realicen a intervalos periódicos de dos a tres años, ya que el control químico debe basarse en el conocimiento de la efectividad biológica de insecticidas y estudios de monitoreo de resistencia de la plaga (Cortez et al. 2013). Anualmente, personal del Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias, reúne información sobre productos potenciales para ser utilizados en las aspersiones regionales contra el PAC. Sin embargo, en años recientes el proceso se ha dificultado debido al cuestionamiento de ingredientes activos utilizados, generalmente por el impacto asociado a enemigos naturales y principalmente a polinizadores, además del alto costo de los nuevos productos en el mercado. Esto ha ocasionado que prácticamente se seleccionen dos o tres insecticidas para programar las aplicaciones contra el PAC, lo que limita la adecuada rotación de insecticidas por otro modo de acción, para el manejo de la resistencia a insecticidas (IRAC 2022). El surgimiento en el mercado de los nuevos insecticidas sintéticos y la disponibilidad de productos derivados de fuentes naturales (biorracionales), marcan la oportunidad para realizar evaluaciones de efectividad biológica e incrementar las alternativas de insecticidas para el manejo regional del vector del HLB. El objetivo del presente estudio fue evaluar la efectividad biológica de insecticidas sintéticos y naturales sobre ninfas del PAC.

Materiales y Método

La evaluación de los insecticidas sobre ninfas de *D. citri* se realizó en una huerta de limón Persa de dos años ($25^{\circ}06'14.346''$ N, $08^{\circ}01'48.2412''$ O), en La Reforma, Angostura, Sinaloa, México, del 01 de junio al 22 de diciembre del 2021. Debido al número de tratamientos (Cuadro 1) las evaluaciones se realizaron en fechas diferentes.

Las aplicaciones se hicieron con una bomba de motor Echo® SHP-800-2 de dos pistolas aspersoras, con boquillas de D-5 doble orificio (aceleración de 200 p.s.i.), en 12 árboles de limón Persa. En cada árbol se aplicaron 500 ml de agua (más acidificante e insecticida) calibrando previamente la aspersora y se procedió a calcular la cantidad correspondiente de cada insecticida utilizada en 12 árboles (288 m^2). Se utilizaron concentraciones al 50, 75, y 100%, siendo esta última, la dosis máxima recomendada en la etiqueta del insecticida comercial. El tratamiento testigo fue asperjado únicamente con agua. Al agua utilizada para la solución de los insecticidas se acondicionó con 1 ml de acidificante/coadyuvante (Surfacid®) por litro de agua, para prevenir la hidrólisis alcalina de los insecticidas y una mejor dispersión

Cuadro 1. Insecticidas Evaluados sobre Ninfas de *Diaphorina citri* en Limón Persa
 Table 1. Insecticides Evaluated on *Diaphorina citri* Nymphs in Persian Lemon

Ingrediente activo	Nombre	Concentración (g IA/ha*)		
		50%	75%	**100%
Ciantraniliprol	Benevia®	20.0	30.0	40.0
Flupyradifurone	Sivanto®	75.0	112.5	150.0
Pymetrozine	Plenum®	187.5	281.3	375.0
Pyriproxifen	Knack®	25.8	38.6	51.5
Tolfenpyrad	Apta®	79.5	119.3	159.0
Spirotetramat	Movento®	37.5	56.3	75.0
Fenpyroximate	Avolant®	12.7	19.1	25.5
Clorantraniliprol	Clorantra®	10.0	15.0	20.0
Flonicamid	Beleaf®	75.0	112.5	150.0
Buprofezin	Applaud®	445.9	668.9	891.9
Extracto de nim	Mix N®	200.0	300.0	400.0
Aceite vegetal	Corion®	1395.0	2092.5	2790.0
<i>Burkholderia</i>	Venerate®	2361.5	3542.3	4723.0
Tierra de diatomeas	Diata-Sin®	1323.0	1984.5	2646.0
Silicio	Diata-Sin®	930.0	1395.0	1860.0
Caolin	Surround®	4750.0	7125.0	9500.0
Sales potásicas	Impide®	683.7	1025.6	1367.4

*g IA/ha (gramos de ingrediente activo / hectarea). **Dosis máxima permitida por aplicación descrita en la etiqueta del insecticida comercial.

en el follaje. Previo a la aplicación de los tratamientos se marcaron con una banda de plástico color amarillo 12 brotes tiernos con presencia de ninfas del PAC, en 12 árboles. Los brotes se confinaron después de la aplicación de los insecticidas en mangas de tela de polipropileno (Agribon®), para aislar a las ninfas del PAC y no quedaran expuestos al efecto de otros factores de mortalidad, principalmente enemigos naturales y factores abióticos.

Los muestreos para determinar la mortalidad se hicieron con una lupa de mano de 20 aumentos a las 48 h después de la aplicación (2 días después de la aplicación de los tratamientos, DAT) y a las 168 h (7 días DAT). El criterio de mortalidad fue ninfas con tonalidad opaca y deshidratada, sin movimiento. Dos y tres semanas DAT, se inspeccionaron otros brotes tiernos presentes en los árboles donde se realizó la aplicación de los insecticidas, para revisar la presencia de infestaciones nuevas del PAC, lo que indicaría la terminación residual de cada tratamiento. Se utilizó un diseño completamente aleatorio con cuatro repeticiones (tres brotes/repetición) y los datos de mortalidad fueron analizados (ANVA), además se realizó una comparación de medias por DMS (Diferencia Mínima Significativa) ($\alpha = 0.05\%$) (Olivares 1994).

Se realizó un estudio del Cociente de Impacto Ambiental (EIQ, por sus siglas en inglés), con un programa del College of Agriculture and Life Sciences (Grant 2010), éste se basa en la metodología del artículo reportado por Kovach et al. (1992), que permite calcular el impacto ambiental de los insecticidas químicos, sobre consumidor, operador (quien realiza la aplicación del insecticida) y organismos benéficos. El EIQ, se realizó únicamente a los tratamientos insecticidas efectivos contra ninfas de *Diaphorina citri*, además de dos insecticidas sintéticos no evaluados en el presente estudio (fenpropatrín: Platino®, concentración 187.5 g IA/ha y

clorpirifos: Clorfos®, concentración 480 g IA/ha), con el objetivo de obtener una referencia de estos productos usados convencionalmente por los citricultores de la región y compararlos con los mencionados anteriormente.

Resultados y Discusión

Diez tratamientos insecticidas evaluados superaron el 30% de mortalidad sobre ninfas de *D. citri* a las 48 h DAT (Fig. 1). *Burkholderia*, el único de origen natural en esta relación, registró mortalidad de 36.9 y 58.3% en dosis del 50 y 100%, respectivamente. Los tratamientos que registraron mayor mortalidad fueron ciantraniliprol dosis 100 y 75%, cuya mortalidad fue de 63 y 64.3%, respectivamente. Los tratamientos de tolfenpyrad dosis 50 y 100%, registraron la menor mortalidad (30 y 30.4%, respectivamente). En el resto de los tratamientos que superaron 30% de mortalidad, ésta osciló entre 31 y 43.5%. Los tratamientos que no se muestran en la tabla, su mortalidad fue menor a 30%. En el análisis estadístico *Burkholderia* y ciantraniliprol (dosis 75 y 100%) no mostraron diferencia significativa entre ellos, pero si con respecto a los demás tratamientos y testigo.

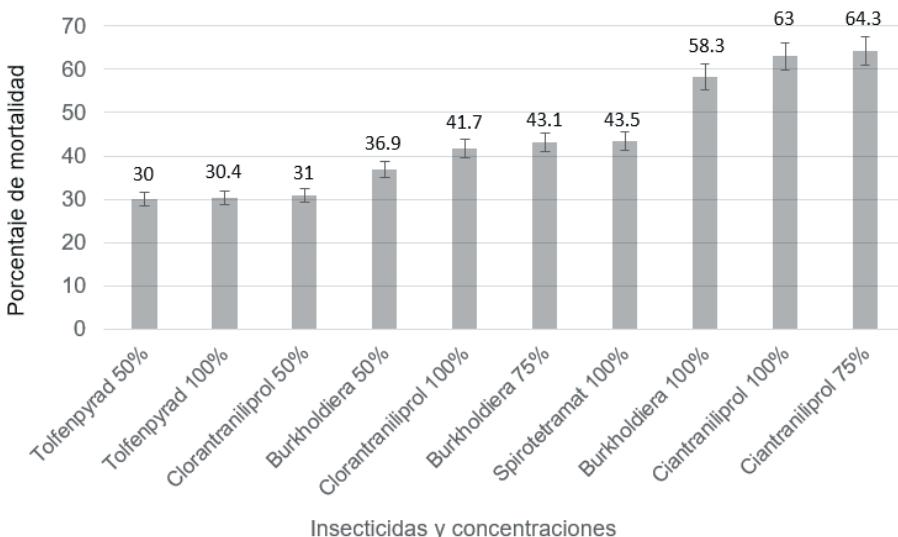


Fig. 1. Insecticidas que superaron 30% de mortalidad en ninfas de *Diaphorina citri* 48 h después de la aplicación.

Fig. 1. Insecticides that exceeded 30% mortality in *Diaphorina citri* nymphs 48 h after application.

Siete insecticidas sintéticos en diferentes concentraciones provocaron arriba de 80% de mortalidad en ninfas del PAC (Fig. 2) 7 días DAT. Spirotetramat, fenpyroximate y clorantraniliprol al 100% produjeron mortalidad de 83.8, 85.1, y 86.6%. Flupyradifurone a 50 y 75% produjo mortalidad de 91.2 y 96.6%. Buprofezin al 75% mató al 92.3% y el tolfenpyrad al 50% mató al 93.1%. Ciantraniliprol y

tolfenpyrad a 75 y 100% mataron 100% de las ninfas del PAC. Flupyradifurone y buprofezin al 100% también mataron a todas las ninfas. Estos tratamientos insecticidas no mostraron diferencia significativa entre ellos, pero si con respecto a los demás tratamientos y testigo.

Flupyradifurone y ciantraniliprol mostraron efecto residual menor de 21 días DAT, pues en la inspección a este intervalo ya se registró presencia del PAC en los brotes tiernos. El resto de los tratamientos insecticidas mostraron un efecto residual menor a 14 días DAT.

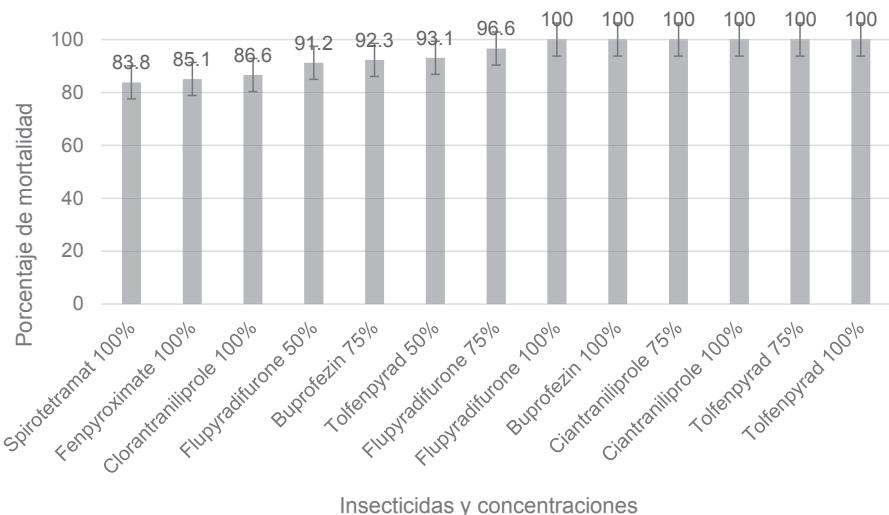


Fig. 2. Insecticidas con efectividad superior a 80% de mortalidad en ninfas de *Diaphorina citri* 7 días después de la aplicación.

Fig. 2. Insecticides with effectiveness greater than 80% mortality in *Diaphorina citri* nymphs 7 days after application.

Existen algunos reportes como el de Miranda et al. (2021) quienes señalan que el tolfenpyrad y el fenpyroximate (ambos a dosis de 1.25 ml/l) obtuvieron mortalidades de 95.65 y 83.87% sobre adultos de *D. citri* en limón mexicano en el Valle de Apatzingán, Michoacán, México. Esta mortalidad es similar a la reportada en el presente estudio, donde el fenpyroximate registró 85% y tolfenpyrad 100%.

Flupyradifurone fue otro tratamiento insecticida que obtuvo mortalidades superiores a 91%. Un estudio realizado por Wen et al. (2021) mencionan que este mismo insecticida puede ser una herramienta nueva y valiosa para los programas de control de *D. citri*, donde puede aumentar su eficiencia al aplicarlo a través del riego por goteo. Otro insecticida con mortalidad superior al 92% fue buprofezin, el cual fue reportado por Tiwari et al. (2012) que suprimió eficazmente la aparición de adultos de *D. citri* y las ninfas chicas (primer-tercer estadio) fueron más susceptibles que las grandes (quinto estadio).

Los tratamientos insecticidas con ≤80% de mortalidad de ninfas del PAC fueron: buprofezin dosis de 50% de concentración (60.8%), fenpyroximate dosis

75% (75%), clorantraniliprol dosis 75% (75.9%), spirotetramat dosis 50% (79.9%), spirotetramat dosis 75% (78.8%), y *Burkholderia* dosis 100% de concentración (79.9%). La mortalidad del resto de los tratamientos fue menor al 60%.

De los insecticidas que superaron 80% de mortalidad fue clorantraniliprol concentración 100% (20 g IA/ha) el que menor EIQ presentó (1.6), con un impacto en el consumidor y operador de 0.6, mientras que el impacto sobre organismos benéficos fue de 3.7 (Cuadro 2). De los que no llegaron al 100% de mortalidad, el que presentó mayor EIQ (46.8) fue buprofezin concentración 75% (668.9 g IA/ha), donde el impacto sobre el operador y el consumidor fue 25.4 y 16.1, respectivamente, mientras que en los organismos benéficos fue 98.9. Por otra parte, de los tratamientos insecticidas que mataron el 100% de las ninfas, el que menor EIQ registró fue ciantraniliprol (EIQ = 3.1) concentración 75% (30 g IA/ha), impacto sobre el consumidor 2.4, operador 2.1, organismos benéficos 6.1, y el que mayor EIQ mostró fue buprofezin (62.4) concentración 100% (891.9 g IA/ha), impacto sobre consumidor y operador fue 33.9 y 21.4, respectivamente, mientras que sobre organismos benéficos fue 131.9.

La EIQ de los insecticidas sintéticos no evaluados en el estudio, pero frecuentemente utilizados por los citricultores de la región, se obtuvo que fenpropatrín a concentración del 100% (187.5 g IA/ha) el EIQ fue 11.3, en consumidor 0.9, operador 2.7, y en organismos benéficos 30.3; mientras que clorpirimifos concentración 100% (480 g IA/ha) el EIQ fue 24. En consumidor 1.8, operador 5.4 y en organismos benéficos 64.7. Lo anterior nos indica que clorpirimifos posee mayor efecto negativo sobre el ambiente, en comparación con fenpropatrín, pero los datos de EIQ no son superiores a los que registró buprofezin en concentraciones de 75 y 100%.

Cuadro 2. Impacto Ambiental de los Tratamientos Insecticidas con Mayor Efectividad sobre Ninfas de *Diaphorina citri*

Table 2. Environmental Impact of the Most Effective Insecticide Treatments on *Diaphorina citri* Nymphs

Tratamiento/Dosis	Mortalidad	EIQ	Impacto sobre			Organismos benéficos
			Consumidor	Operador		
Pirotetramat 100%	83.8%	15.7	2.7	5.8		38.7
Fenpyroximate 100%	85.1%	8.6	1.3	2.7		21.9
Clorantraniliprol 100%	86.6%	1.6	0.6	0.6		3.7
Flupyradifurone 50%	91.2%	9.6	6.0	3.3		19.4
Flupyradifurone 75%	96.6%	14.4	9.0	5.0		29.1
Flupyradifurone 100%	100.0%	19.2	12.0	6.7		38.8
Buprofezin 75%	92.3%	46.8	25.4	16.1		98.9
Buprofezin 100%	100.0%	62.4	33.9	21.4		131.9
Ciantraniloprol 75%	100.0%	3.1	2.4	1.6		4.6
Ciantraniliprol 100%	100.0%	4.2	3.2	2.1		6.1
*Tolfenpyrad 50%	93.1%	11.9	1.7	6.2		27.7
*Tolfenpyrad 75%	100.0%	17.8	2.6	9.2		41.5
*Tolfenpyrad 100%	100.0%	23.7	3.5	12.3		55.4

*En la base de datos para calcular el impacto de los insecticidas no se encontró tolfenpyrad, en cambio se muestran los datos del tebunferpyrad, un insecticida muy similar, con el mismo modo y sitio de acción que tolfenpyrad (IRAC 2022).

De los tratamientos insecticidas que superaron el 80% de mortalidad, el que presentó menor costo fue clorantraniliprol con un precio de \$29.00 a 20 g IA/ha, y el de mayor costo (\$100.00 US) fue buprofezin a 891.9 g IA/ha. Con el insecticida ciantraniliprol a concentraciones de 30 y 40 g IA/ha con 100% de mortalidad, se obtiene un ahorro de \$17.70 US con la dosis baja. De la misma manera, tolfenpyrad utilizando una menor concentración se obtiene un ahorro de \$16.10 US, debido a que las concentraciones de 119.3 y 159 g IA/ha mataron a todas las ninfas.

Los insecticidas de origen natural, extracto de nim, aceite vegetal, sales potásicas, tierra de diatomeas (silicio), y caolín no fueron efectivos. La bacteria *Burkholderia* mostró resultados moderados de mortalidad a las 48 h DAT, y a la concentración de la dosis alta de 4723 g IA/ha (100%) 7 d DAT, la mortalidad fue casi del 80%, por lo tanto, puede considerarse una alternativa interesante para el control del PAC, en sistemas de producción orgánica, pues además tiene registro por el Instituto de Revisión de Materiales Orgánicos (OMRI por sus siglas en inglés), sin embargo posee elevado costo económico de \$120.00 US, en la dosis indicada.

Cuadro 3. Costo (U.S. Dólares) de la Dosis Insecticida Evaluada Contra Ninfas de *D. citri*

Table 3. Cost (U.S. Dollars) of the Insecticide Dose Evaluated Against Nymphs of *D. citri*

Insecticida	g IA/ha	Mortalidad	Costo dosis/ha
Spirotetramat	75.0	83.8%	87.50
Fenpyroximate	25.5	85.1%	150.40
Clorantraniliprol	20.0	86.6%	29.00
Flupyradifurone	75.0	91.2%	35.80
Buprofezin	38.6	92.3%	75.00
Tolfenpyrad	79.5	93.1%	32.20
Flupyradifurone	112.5	96.6%	53.70
Flupyradifurone	150.0	100.0%	71.60
Buprofezin	891.9	100.0%	100.00
Ciantraniliprol	30.0	100.0%	53.30
Ciantraniliprol	40.0	100.0%	71.00
Tolfenpyrad	119.3	100.0%	48.20
Tolfenpyrad	159.0	100.0%	64.30

Los siete insecticidas con elevada efectividad biológica sobre *D. citri* (spirotetramat MoA 23, clorantraniliprol MoA 28, ciantraniliprol MoA 28, flupyradifurone MoA 4-D, buprofezin MoA 16, tolftenpyrad MoA 21A, fenpyroximate MoA 21A; IRAC 2022), se agrupan en cinco diferentes modos y sitios de acción (IRAC 2022), pues el clorantraniliprol y el ciantraniliprol, y aparte el tolftenpyrad y el fenpyroximate comparten características y se ubican juntos; lo que debe ser considerado para el manejo preventivo de la resistencia a insecticidas (IRAC 2022), considerando además la fenología del cultivo, la presencia de otros insectos plaga y organismos no blanco (IRAC 2016), como enemigos naturales e insectos polinizadores, lo cual ha sido mencionado por Étienne et al. (1998), Grafton-Cardwell et al. (2021), Michaud (2004), Monzo et al. (2014) entre otros.

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Referencias Citadas

- Boina, D. R., y J. R. Bloomquist. 2015. Chemical control of the Asian citrus psyllid and of huanglongbing disease in citrus. Pest. Manag. Sci. 71: 808-823. DOI: 10.1002/ps.3957.
- Chen, J., X. Pu, X. Deng, S. Liu, H. Li, and E. Civerolo. 2009. A phytoplasma related to "Candidatus Phytoplasma asteris" detected in citrus showing Huanglongbing (Yellow Shoot Disease) symptoms in Guangdong, P. R. China. Phytopatol. 99: 236-242.
- Cortez, M. E., J. Pérez, V. González, y H. Medina. 2011. Fluctuación poblacional de *Diaphorina citri* Kuwayama e insecticidas biorracionales para su control en cítricos en Sinaloa. Informe final de proyecto de investigación. INIFAP-CIRNO-CEVAF. Juan José Ríos, Sinaloa.
- Cortez, M. E., J. Loera, L. Hernández, J. Barrera, A. Fontes, U. Díaz, J. Jasso, M. Reyes, M. Manzanilla, y J. López. 2013. Manual para el uso de insecticidas convencionales y alternativos en el manejo de *Diaphorina citri* Kuwayama en cítricos, en México. SAGARPA-INIFAP. Folleto Técnico Núm. 37. ISBN: 978-607-37-0016-0. Juan José Ríos, Sinaloa, México.
- Della, V. J., D. de Andrade, R. Gomes, y M. da Costa, F. 2018. Effects of insecticide and acaricide mixtures on *Diaphorina citri* control. Rev. Bras. Frutic. 12: 1-7. doi.org/10.1590/0100-29452019076.
- Etienne, J., D. Burckhard, y C. Grapin. 1998. *Diaphorina citri* (Kuwayama) en Guadalupe, premier signalement pour les Caraïbes (Hem.: Psyllidae). Bull. Soc. Entomol. Fr. 103: 32.
- Grafton-Cardwell, E., R. Baldwin, J. Becker, A. Eskalen, C. Lovatt, S. Rios, J. Adaskaveg, B. Faber, D. Haviland, K. Hembree, J. Morse, and B. Westerdahl. 2021. UC IPM Pest Management Guidelines: Citrus. Revised continuously. UC ANR Publication 3441. Oakland, CA.
- Grant, J. A. 2010. Calculator for Field Use EIQ (Environmental Impact Quotient). New York State Integrated Pest Management Program, Cornell Cooperative Extension, Cornell University. <https://nysipm.cornell.edu/eiq/calculator-field-use-eiq/>
- IRAC (Insecticide Resistance Action Committee. 2016. The Asian citrus psyllid, *Diaphorina citri*: 'Insecticide Resistance Management' is the Basis of a Successful IPM Program. IRAC Sucking Pest Team. Poster version 3.3. <https://irac-online.org/pests/diaphorina-citri/posters/>
- IRAC (Insecticide Resistance Action Committee). 2022. IRAC Mode of Action Classification Scheme. Issued, June 2022; Version 10.3. <https://irac-online.org/mode-of-action/>
- Iqbal, J., H. Hussain, M. Latif, M. Baig, A. Owayss, S. Raweh, and S. Alqarni. 2020. A field study investigating the insecticidal efficacy against *Diaphorina citri* Kuwayama on Kinnow mandarin, *Citrus reticulata* Blanco trees. Saudi J. Biol. Sci. 27: 1237-1241. <https://doi.org/10.1016/j.sjbs2020.02.006>.

- Kovach, J., C. Petzoldt, J. Degni, and J. Tette. 1992. A method to measure the environmental impact of pesticides. New York's Food and Life Sciences. Bulletin 139: 1-8.
- Olivares, S. E. 1994. Paquete de diseños experimentales. FAUANL. Versión 2.5. Facultad de Agronomía, UANL. Marín, N. L.
- Michaud, J. P. 2004. Natural mortality of Asian citrus psyllid (Homoptera: Psyllidae) in central Florida. Biol. Control 29: 260-269.
- Miranda, R. C., S. Perales, S. Miranda, y M. Miranda. 2021. Insecticidas de bajo impacto ambiental para el control de *Diaphorina citri* Kuwayama, 1908 (Hemiptera: Liviidae) en limón mexicano (*Citrus aurantifolia* (Christm.) Swingle). Rev. Chilena Entom. 47: 723-732. <https://www.scielo.cl/pdf/rche/v47n4/0718-8994-rche-47-04-723.pdf>
- Monzo, C., J. A. Qureshi, and P. A. Stansly. 2014. Insecticide sprays, natural enemy assemblages and predation on Asian citrus psyllid, *Diaphorina citri* (Hemiptera: Psyllidae). Bull. Entomol. Res. 104: 576-585.
- SENASICA (Servicio Nacional de Sanidad, Inocuidad y Calidad Agroalimentaria). 2009. <http://www.senasica.gob.mx>
- Tiwari, S., J. Clayson, E. Kuhns, and L. Stelinski. 2012. Effects of buprofezin and diflubenzuron on various developmental stages of Asian citrus psyllid, *Diaphorina citri*. Pest Manag. Sci. 68: 1405-1412.
- Wen, Y., H. Meng, C. Zhao, F. Lin, and H. Xu. 2021. Evaluation of flupyradifurone for the management of the Asian citrus psyllid *Diaphorina citri* via dripping irrigation systems. Pest Manag. Sci. 77: 2584-2590.

Descripción de *Mesoamericaleyrodes* gen. nov.¹ y Dos Especies de Moscas Blancas de México**Description of *Mesoamericaleyrodes* gen. nov.¹ and Two Species of Whiteflies from Mexico**

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Resumen. El género *Mesoamericaleyrodes* Carapia-Ruiz gen. nov. es descrito, incluyendo la especie *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov. encontrada en Tepic, Nayarit sobre el envés de las hojas de *Lonchocarpus* (Papilionaceae), y *M. michoacan* Carapia-Ruiz sp. nov. hallada en Pichilingillos, Michoacán en el envés de las hojas de *Piscidia grandiflora* (Donn.Sm.) S.F. Bake (Papilionaceae). Se ilustra con microfotografías de estructuras morfológicas de puparías del género y especies. Se discute la separación con grupos cercanos.

Abstract. The genus *Mesoamericaleyrodes* Carapia-Ruiz gen. nov. is described, including the species *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov. found at Tepic, Nayarit on the underside of *Lonchocarpus* (Papilionaceae) leaves, and *M. michoacan* Carapia-Ruiz sp. nov. found at Pichilingillos, Michoacan, on the underside of *Piscidia grandiflora* (Donn.Sm.) S.F. Bake (Papilionaceae) leaves. Microphotographs of morphological structures of the genus and species puparia are provided. Separation from other near groups is discussed.

Introducción

En estudios de moscas blancas de la subfamilia Aleyrodinae de México, Baker (1937) reporta los géneros *Aleurocanthus*, *Aleuroplatus*, *Aleurothrixis*, *Aleurotrachelus*, *Aleyrodes*, *Bermisia*, *Dialeurodes*, *Singhiella* (*S. Citrifolii*), *Tetralecia* (*Aleuropleurocelus*), *Tetraleurodes*, y *Trialeurodes*. Sampson y Drews (1941) reportan a *Aleuroglandulus* y *Pealius*, y describen a *Paraleurolobus*. Russell (1947) reporta a *Aleuroparadoxus* *Aleurothithius*, y *Aleurocerus* Russell (1986). *Aleurocybotus* es reportado por Vejar-Cota et al. (2009). Carapia-Ruiz et al. (2009) describen el género *Cuautlaleyrodes*, mientras que *Parabemisia* y *Siphoninus* son mencionados por Carapia Ruiz y Sánchez Flores (2018), Sánchez-Flores et al. (2021) reportaron *Filicaleyrodes*. En Nayarit y Michoacán México fueron encontrados dos especies congéneres sobre dos Papilionaceae que no corresponde a alguno de los géneros conocidos, por lo que se realizó el presente estudio con la finalidad de describir y clasificar este género y sus dos especies.

¹Hemiptera: Aleyrodidae, Aleyrodinae

Materiales y Metodos

En abril 2004 en Tepic, Nayarit, México se colectaron especímenes de una especie de mosca blanca sobre *Lonchocarpus* (Papilionaceae), y en junio del 2006 otra especie fue colectada en un árbol del género *Piscidia grandiflora* (Donn.Sm.) S.F. Bake (Papilionaceae), en Pichilinguillos Michoacán. La metodología para la preparación de especímenes fue de Martin (2004)

Resultados y Discusión

Descripcion. *Mesoamericaleyrodes* Carapia-Ruiz gen. nov. (Figs. 1-8)

Forma general. Pupas blanco-amarillentas comúnmente con áreas oscuras en el submargen, y en la parte media del cuerpo, con filamentos de cera, similar a algunos grupos de *Trialeurodes*.

Especímenes en portaobjetos. Cuerpo elíptico-oval cercano a 1 mm de largo.

Margen y submargen. Margen provisto de granulaciones, submargen no separado del disco dorsal el cual presenta papilas normalmente arregladas en una hilera ampliamente espaciada acompañadas de poros y poretes. Setas marginal anterior y marginal posterior presentes y relativamente pequeñas.

Dorso. Cefalotórax. Superficie uniforme provista de papilas muy aparentes y variando en tamaño y número en algunas partes puede presentar depresiones, setas céfálicas presentes, mesotorácicas y metatorácicas ausentes. Sutura longitudinal de la muda aproximadamente la mitad de la longitud del cuerpo, sutura transversa de la muda curva en forma de W alcanzando el margen o el submargen del cuerpo al nivel de la sutura metatorácica o posterior a esta.

Abdomen. Segmentos abdominales I-VIII claramente visibles en la parte media y su longitud solo varía ligeramente entre ellos. Depresiones abdominales presentes con forma y tamaño variable entre las especies, área del disco dorsal con papilas evidentes de todos los segmentos abdominales, la parte media de los segmentos abdominales con dos hileras de papilas.

Orificio vasiforme. Alargado longitudinalmente y de forma semitriangular, opérculo truncado semitrapezoidal cubre solo cerca de la mitad anterior de la língula, la cual es alargada y espatulada con ligeros lóbulos en la mitad posterior, incluida en el orificio vasiforme, este no definido anteriormente. Longitud del orificio vasiforme varía ligeramente con respecto a la distancia de este al margen posterior y también a la sutura abdominal VIII. Setas abdominales VIII anterolaterales al orificio vasiforme bien definidas.

Vientre. Antenas se extienden posteriormente al par de las patas protorácticas, patas mesotóracicas, patas metatóracicas con su cutícula aparentemente lisa como en el resto parte ventral, un par de sacos adhesivos cerca de la base del primer par de patas, cutícula abdominal lisa.

Quetotaxia. Con setas marginales anteriores, céfálicas, abdominales 1, abdominales VIII, caudales, marginales posteriores. Setas mesotorácicas y metatorácicas ausentes.

Etimología. *Mesoamericaleyrodes* Carapia-Ruiz gen. nov. es referido a Mesoamérica, región donde se encontraron los especímenes.

Distribución. Nayarit y Michoacán, México.

Comentarios Taxonómicos. *Mesoamericaleyrodes* Carapia-Ruiz gen. nov. es un género que se describe de dos especies de México, sus características lo

permiten incluir en la tribu Bemisini por tener orificio vasiforme triangular, língula espatulada no presentan pliegues, peines, poros traqueales torácicos o abdominales.

Especie tipo. *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov.

Descripción. *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov.

Forma general. Pupas blanco-amarillentas algunas veces con áreas oscuras en el submargen, con filamentos de cera en el dorso, similar a algunos grupos de *Trialeurodes*.

Especímenes en portaobjetos. Cuerpo elíptico-oval de 1.0–1.3 mm de largo y 0.8 a 1 mm de ancho (Fig. 1).

Margen y submargen. Margen con débiles granulaciones, submargen no separado del disco dorsal el cual presenta papilas normalmente arregladas en una hilera ampliamente espaciada acompañadas de poros y poretes (Fig. 1a), el summargen con pigmento marrón oscuro, variando entre los especímenes. Setas marginal anterior y marginal posterior presentes y relativamente pequeñas (Fig. 4).

Dorso. Cefalotórax. Superficie en su mayor parte lisa provista de papilas muy distintivas y variando en tamaño de 10-15 µm de ancho, el número aproximado de pares de papilas es 20-25 en el submargen, 12-14 en la región submediana y 35-38 en el subdorso; presenta dos a tres depresiones por segmento en la parte submediana, setas céfálicas presentes, mesotorácicas y metatorácicas ausentes. Sutura longitudinal de la muda de 520 µm de largo, sutura transversal de la muda se extiende en forma curva y termina en el margen pupal, aproximadamente el nivel de la sutura metatorácica (Figs. 1, 2).

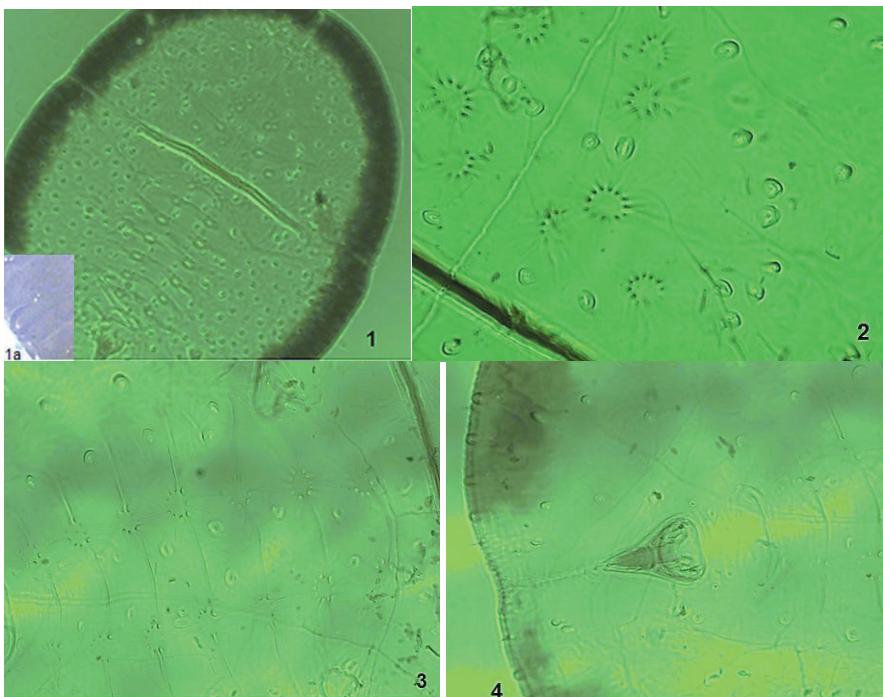
Abdomen. Segmentos abdominales I-VIII claramente visibles en la parte media con longitud aproximada de: segmento I 51 µm, segmento II 52 µm, segmento III 51 µm, segmento IV 51 µm, segmento V 50 µm, segmento VI 48 µm, segmento VII 50.5 µm, y segmento VIII 51.5 µm. Depresiones abdominales un par por segmento de 21-30 µm de ancho. Papilas evidentes de 10-15 µm en los segmentos abdominales, aproximadamente el número de pares papilas es 12-14 submedianas, 35-40 subdorsales, y 28-35 submarginales. Surco caudal angosto bien definido (Fig. 3).

Orificio vasiforme. Alargado longitudinalmente semitriangular de 78-82 µm largo por 65-70 µm en la parte más ancha; opérculo truncado semitrapezoidal 45-47 µm de largo por 55-57 µm de ancho, cubre completamente la mitad anterior de la língula; língula de 70-75 µm largo y 5.6 µm de ancho, terminando en la parte posterior de del orificio vasiforme; Distancia del margen posterior del orificio vasiforme al margen pupal 70 µm, surco caudal angosto bien definido (Fig. 4).

Vientre. Antenas se extienden posteriormente al par de las patas protorácticas, patas sin una banda ancha irregular de espínulas, cutícula torácica aparentemente lisa en el resto parte ventral, un par de sacos adhesivos cerca de la base del primer par de patas, cutícula abdominal lisa. Octavo segmento abdominal con un par de espiráculos y un par de setas ligeramente anterior a los espiráculos

Quetotaxia. Largo de las setas: Setas marginales anteriores 10-12 µm, setas céfálicas 7-15 µm, mesotorácicas y metatorácicas ausentes, setas de segmento abdominal I de 10-25 µm, setas de segmento abdominal VIII de 6-55 µm, setas caudales de 7-35 µm, setas marginales posteriores de 10-12 µm.

Material tipo: Holotipo: Tepic, Nayarit, México, en el envés de hojas *Lonchocarpus* (Papilionaceae). 8-abril-2004. Colector: Vicente Emilio Carapia-Ruiz, depositado en la Colección Nacional de Insecto CNI del instituto de Biología de la UNAM, Ciudad de México, México. Paratipos, 22, Mismos datos del Holotipo, dos



Figs. 1-4. *Mesoamericaleyrodes nayarit* sp. nov. 1) Pupario, 2) Cefalotorax, 3) Abdomen, 4) Orificio vasiforme.

Figs. 1-4. *Mesoamericaleyrodes nayarit* sp. nov. 1) Puparium, 2) Cephalothorax, 3) Abdomen, 4) Vasiform orifice.

depositados en la colección CNI, 18 depositados en la colección del autor VECR y dos en la colección OASF.

Etimología. *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov. es referido al estado de Nayarit, México, lugar donde se encontraron los especímenes.

Comentarios Taxonómicos. *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov. es una especie que se describe de Tepic, Nayarit, México, sus características distintivas son: por su forma semicircular-oval, papillas dorsales relativamente pequeñas (10-15 µm), submargen con una hilera subm marginal de poros y poretes, surco caudal bien definido, diferiendo de *M. nayarit* sp. nov. por presentar forma marcadamente alargada elíptica, las papillas dorsales relativamente más grandes (17-21 µm). El surco caudal pobremente definido, y no presenta hilera subm marginal de poros y poretes,

Hospederos. *Lonchocarpus* (Papilionaceae).

Distribución. *Mesoamericaleyrodes nayarit* Carapia-Ruiz sp. nov. se encontró en Tepic Nayarit, México

Descripción. *Mesoamericaleyrodes michoacan* Carapia-Ruiz sp. nov.

Forma general. Pupas blanco-amarillentas, normalmente con áreas oscuras en el submargen y parte media del cuerpo, con filamentos de cera en el dorso, similar a algunos grupos de *Trialeurodes*.

Especímenes en portaobjetos. Cuerpo elíptico-oval de 900–940 µm de largo y 590–620 µm de ancho (Fig. 5).

Margen y submargen. Margen con granulaciones, submargen no separado del disco dorsal el cual presenta papilas en una hilera ampliamente espaciada, normalmente con áreas oscuras en el submargen, surcos submarginales definidos. Setas marginal anterior y marginal posterior presentes y relativamente pequeñas (Fig. 6).

Dorso. Cefalotórax. Superficie en su mayor parte lisa provista de papilas prominentes variando en tamaño de 17-21 µm de ancho, 12-13 pares en el área submediana, 16-19 pares en la parte subdorsal y 10-12 pares en el submargen; en algunas partes puede presentar depresiones, normalmente de dos depresiones en la parte media del metatórax, tres en el mesotórax y tres en el protórax, comúnmente una de estas depresiones está cerca a la sutura intersegmental, setas cefálicas presentes, mesotorácicas y metatorácicas ausentes. Sutura longitudinal de la muda de 430 µm de largo, sutura transversal de la muda se extiende en forma curva y termina en el margen pupal (Fig. 6).

Abdomen. Segmentos abdominales I-VIII claramente visibles en la parte media con longitud aproximada de: segmento I 45 µm, segmento II 45 µm, segmento III 43 µm, segmento IV 43 µm, segmento V 42 µm, segmento VI 38 µm, segmento VII 36 µm, y segmento VIII 50 µm. Un par de depresiones abdominales medianas por segmento de 20-22 µm de ancho; papillas prominentes de 16-18 µm de ancho, el número de pares de papillas es de 12-13 submedianas, 18-20 subdorsales, y 12-14 submarginales (Figs. 5, 7).

Orificio vasiforme. Alargado semitriangular de 80 µm largo por 58 µm en la parte más ancha; Opérculo truncado 25-27 µm de largo por 40-43 µm de ancho, cubre la mitad anterior de la língula; língula de 40-42 µm largo y 9-11 µm de ancho; Distancia del margen posterior del orificio vasiforme al margen aparente 64 µm (Fig. 8).

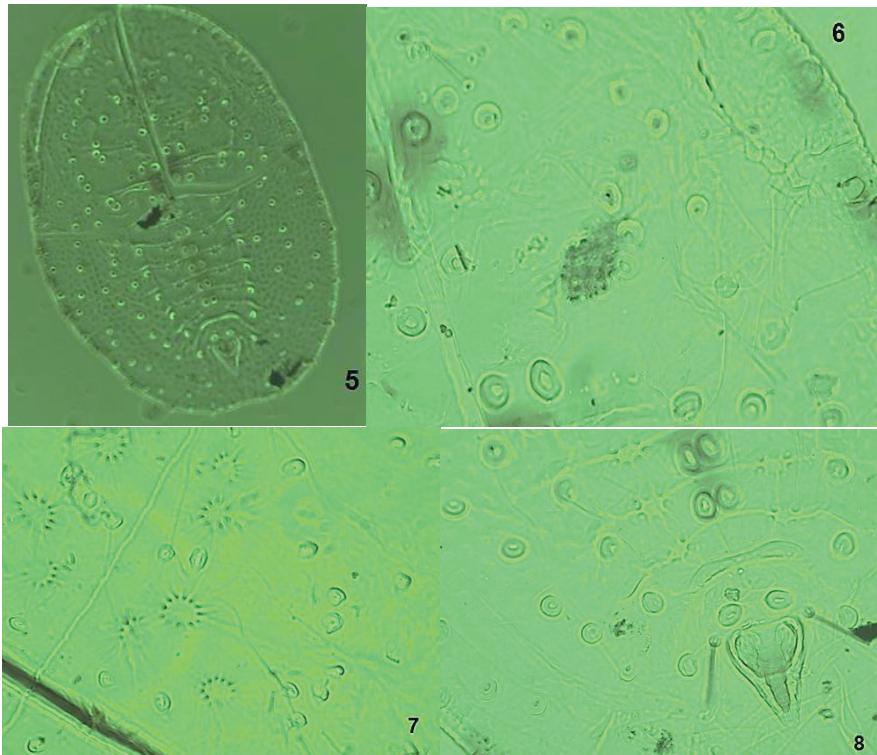
Vientre. Antenas se extienden posteriormente al par de las patas protorácicas, patas sin espínulas, cutícula aparentemente lisa en el resto parte ventral, un par de sacos adhesivos cerca de la base del primer par de patas. Octavo segmento abdominal con un par de espiráculos y un par de setas ligeramente anteriores a los espiráculos.

Quetotaxia. Largo de las setas: marginales anteriores 9-10 µm, cefálicas 15-28 µm, mesotorácicas y metatorácicas ausentes, setas de segmento abdominal I de 28 µm, setas de segmento abdominal VIII de 30 µm, caudales de 50 µm, marginales posteriores de 9-10 µm.

Material tipo. Holotipo: Pichilinguillos, Michoacán, México, en el envés de hojas de *Piscidia grandiflora* 18-junio-2006. Colector: Vicente Emilio Carapia-Ruiz depositado en la Colección Nacional de Insectos CNI del Instituto de Biología de la UNAM, Ciudad de México. Paratipos, 20, Mismos datos del Holotipo, 18 depositados en la colección del autor y dos en la colección OASF.

Etimología. *Mesoamericaleyrodes michoacan* Carapia-Ruiz sp. nov. es referido al Estado de Michoacán, México.

Comentarios taxonómicos. *Mesoamericaleyrodes michoacan* Carapia-Ruiz sp. nov. es una especie que se describe de Michoacán, México, sus características distintivas son: forma marcadamente alargada elíptica, las papillas dorsales relativamente grandes (17-21 µm). El surco caudal pobremente definido, y no presenta hilera submarginal de poros y poretes que difieren de *M. nayarit* sp nov. como sea indicó anteriormente.



Figs. 5-8. *Mesoamericaleyrodes michoacan* sp. nov. 5) Pupario, 6) Cefalotórax, 7) Abdomen, 8) Orificio vasiforme.

Figs. 5-8. *Mesoamericaleyrodes michoacan* sp. nov. 5) Puparium, 6) Cephalothorax, 7) Abdomen, 8) Vasiform orifice.

Distribución. Pichilinguillos Michoacán, México.

Hospederos. *Piscidia grandiflora* (Papilionaceae).

Agradecimiento

A J. H. Martin Por los comentarios sobre este género y su gran aportación de moscas blancas de Belice.

Referencias Citadas

- Baker, J. M. 1937. Notes on some Mexican Aleyrodidae. An. Inst. Biol. Univ. Mex. 8: 599-629.
- Carapia-Ruiz, V. E., A. Castillo-Gutiérrez, Y. Ortega-Saad, V. M. Hernández-Velásquez, G. Peña-Chora, y M. E. Nuñez-Valdez. 2009. Description of *Cuautlaleyrodes canis* gen. et sp. nov. of whiteflies (Hemiptera:Aleyrodidae) from Mexico. TecnoINTELECTO 6: 2-7.

- Carapia-Ruiz, V. E., y O. A. Sánchez-Flores. 2018. Moscas blancas (Hemiptera: Aleyrodidae) de México. Entomol. Mex. 5: 616-621.
- Martin, J. H. 2004. Whiteflies of Belize (Homoptera: Aleyrodidae) Part 1-Introduction and account of the subfamily Aleurodicinae Quaintance y Baker. Zootaxa 681: 1-119.
- Russell, L. M. 1947. A classification of the whiteflies of the new tribe Trialeurodini (Homoptera: Aleyrodidae). Rev. Entomol. (R. De J.) 18: 1-44.
- Russell, L. M. 1986. The whitefly genus *Aleurocerus* Bondar (Hemiptera, Homoptera, Aleyrodidae). Entomography 4: 137-183.
- Sampson, W. W., and E. A. Drews. 1941. Fauna Mexicana IV. A review of the Aleyrodidae of México. An. Esc. Nac. Cien. Biol. (México) 2: 143-189.
- Sánchez-Flores O. A., V. E. Carapia-Ruiz, A. K. Dubey, and A. Castillo-Gutiérrez. 2021. *Filicaleyrodes huastecus* sp. nov. from the Western Hemisphere with a Key to Species. Southwest. Entomol. 46: 451-456.
- Vejar-Cota, G., L. D. Ortega-Arenas, y V. E. Carapia-Ruiz. 2009. Primer registro de la mosca blanca de los cereales *Aleurocybotus occidus* Russell (Hemiptera: Aleyrodidae) y su impacto potencial como plaga de gramíneas en el norte de Sinaloa. Acta Zool. Mex. (n.s.) 5: 33-48.

Insectos y Ácaros Fitófagos Asociados con la Guanábana *Annona muricata* L.: Una Revisión**A Review of Insect and Mite Pests of Soursop *Annona muricata* L.**

Luis Martín Hernández Fuentes^{1*}, José Joaquín Velázquez Monreal², Héctor González Hernández³, Carlos Patricio Illescas Riquelme⁴, y Miguel Ángel Manzanilla Ramírez²

Resumen. Nativa de América Central, la guanábana, *Annona muricata* L., se cultiva principalmente en la región tropical de América. Con el incremento en superficie e importancia económica, su cultivo presenta limitantes principalmente de plagas y enfermedades. El objetivo es reunir la información disponible para integrar un mejor manejo de plagas en este cultivo, se presenta un listado de insectos y ácaros asociados con el cultivo. Solo se incluyen las especies en asociación con el cultivo por observación directa o emergencia del adulto en los insectos holometábolos de alguna estructura de la planta. Se mencionan las especies de insectos y ácaros cuya identificación taxonómica se revisó por especialistas, consultas en base de datos y/o uso de claves. Se obtuvieron 116 especies de insectos y dos de ácaros en asociación alimenticia con *A. muricata*. Se presentan ocho órdenes de insectos con 33 familias y uno de ácaros con dos familias. Destaca el orden Hemiptera con 14 familias y 69 especies. Existe mayor cantidad de insectos barrenadores, masticadores y chupadores de frutos (57%) en comparación con otras estructuras de la planta. El insecto de mayor importancia es *Bephratelloides* por los daños que causa y su amplia distribución. Le sigue en importancia *Cerconota anonella* Sepp distribuida principalmente en Centro y Sudamérica. Predominan los adultos con 75% causando daños. El 50% de las plagas mencionadas dañan solo una estructura del cultivo (frutos) no obstante, creemos que se requieren mayores observaciones en este sentido. Especies que no tienen amplia distribución como *Optatus palmaris* Pascoe y *Maconellicoccus hirsutus* Green pueden causar graves daños en el cultivo. La distribución de las especies plaga es tropical y subtropical en asociación con la distribución y origen de la guanábana.

Abstract. Native to Central America, soursop, *Annona muricata* L., is cultivated mainly in the tropical region of the Americas. With the increase in surface area and economic importance, its cultivation has limitations mainly due to pests and diseases. We summarized available information to integrate a better pest management in this crop, creating a list of insects and mites associated with the crop. Only species in

¹Km. 6, Entronque a Santiago Ixquintla, Carretera Internacional México-Nogales, Santiago Ixquintla, Nayarit. C. P. 63300.

²Km. 35 Carretera Colima-Manzanillo, Tecomán, Colima. C.P. 28100.

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association with cultivation by direct observation or adult emergence in holometabolous insects from some plant structures are included. Insects and mites whose taxonomic identification was reviewed by specialists, database queries, and/or use of keys are mentioned. In total, 116 species of insects and two of mites are recorded associated with *A. muricata*, in eight orders of insects with 33 families and one of mites with two families. Hemiptera had 14 families and 69 species. There were more boring, chewing, and fruit-sucking insects (57%) than on other plant structures. The most important insect was *Bephratelloides* because of damage it causes and its wide distribution. It was followed in importance by *Cerconota anonella* Sepp, distributed mainly in Central and South America. Adults dominate with 75% of damage. Fifty percent of the pests damaged only fruits, but we believe more observations are required. Species that do not have a wide distribution such as borer weevil, *Optatus palmaris* Pascoe, and pink hibiscus mealybug, *Maconellicoccus hirsutus* Green, can cause serious damage to the crop.

Introducción

De las especies de *Annona* que presentan frutos comestibles, la guanábana *A. muricata* L., actualmente cuenta con mayor desarrollo agronómico y superficie establecida. En México, se producen anualmente 30,790 toneladas cuyo valor asciende a 248.2 millones de pesos (SIAP 2019). Es nativa de América Central (Laca y Ramon 1999, Prasad et al. 2014) y se cultiva principalmente en México, Venezuela, Ecuador, Colombia, y Brasil en áreas con un clima cálido desde el nivel del mar hasta una altitud de 800 m. México y Brasil son los principales productores con 8,000 ha (Lemos 2011). Se presenta un listado de insectos y ácaros asociados con el cultivo. Se revisaron las bases de datos de ResearchGate (<https://www.researchgate.net>), CABI Invasive Species Compendium, Crop Protection Compendium, Plantwise Knowledge Bank (<https://www.cabi.org/>), y Scopus (<https://www.scopus.com>). Únicamente se enlistaron las especies provenientes de reportes donde se corroboró u observó 1.- emergencia de adultos holometábolos donde el estado larvario se desarrolló dentro del tejido vegetal, 2.- observación directa de daños de al menos un estado de desarrollo del insecto o ácaro, 3.- colonias de hemimetábolos sésiles de diferentes estados de desarrollo habitando una o varias estructuras vegetales de *A. muricata*, 4.- reportes de asociación e identificación de insectos y ácaros fitófagos con *A. muricata* cuya identidad se corroboró por especialistas en los grupos taxonómicos respectivos, y/o utilización de claves taxonómicas específicas. Se descartaron los reportes que no mencionan la especie y donde no se verificó la asociación fitófaga, debido a que existe la probabilidad de que el artrópodo, aún y cuando sea fitófago, podría estar al momento de la recolecta u observación en alguna estructura vegetal de *A. muricata* en estado de reposo, resguardo, pernocta, apareamiento o forma parte de su nicho, entendiéndose a éste como el papel que desempeña la especie en la comunidad en relación con los factores bióticos y abióticos (Elton 1927), sin ser necesariamente su hospedante. O bien como lo señalan Aluja et al. (1987), en el caso de hospedantes de moscas de la fruta del género *Anastrepha*, algunos reportes en la literatura podrían ser el resultado de errores en la identificación y muestreo (SIC). Algunos nombres específicos cambiaron respecto a la publicación original debido a las actualizaciones taxonómicas.

Se reportan 117 especies de insectos y dos de ácaros en asociación alimenticia con *A. muricata* (Tabla 1), las cuales se encuentran distribuidos en países

Tabla 1. Insectos y Ácaros Fitófagos Asociados con *Annona muricata*
 Table 1. Insects and Mites Phytophagous Related with *Annona muricata*

Familia	Especie	Estado biológico	Daños	País	Referencia*
Eurytomidae	<i>Bephratelloides cubensis</i>	a,b	3	México Puerto Rico Estados Unidos Honduras Cuba	1-4 5 6 7 8
	<i>B. limai</i> Bondar	a,b	3	Brasil	9-10
	<i>B. maculicollis</i> Cameron	a,b	3	Venezuela	11
				Trinidad y Tobago	12
				Puerto Rico	13
				Costa Rica	14
Apidae	<i>B. pomorum</i> Fabricius	a	2-3	Brasil	15
	<i>Trigona spinipes</i> Fabricius	b	2-3	Brasil	16
Depressariidae	<i>Cerconota anonella</i> Sepp	b	3	Brasil México Brasil Venezuela	10 2-3 9-10,16-18 11
				Trinidad y Tobago	12
Erebidae	<i>Gonodonta pyrgo</i> Cramer	b	1-3	Colombia	19
	<i>G. fulvanguilata</i> Cramer	b	1	Honduras	7
Lycaenidae	<i>Thecla orteignus</i> Cramer	b	2-3	Costa Rica	14
Pyralidae	<i>Elasmopalpus lignosellus</i> Zeller	b	4	México	4,20-21
	<i>Cocytius antaeus</i> Drury	b	1	Brasil	16
	<i>C. duponchel</i> Poey	b	1	México	3
				Costa Rica	14
Sphingidae	<i>Protomabulix stringilis</i> Clark	b	1	Puerto Rico	5
Papilionidae	<i>Graphium eurypylus</i> L.	b	1	Australia	11
	<i>G. agamemnon</i> L.	b	1	Vietnam	5
				Brasil	22
Tephritidae	<i>Bactrocera invadens</i> Drew, Tsuda & White	b	3	Tanzania	23
	<i>B. dorsalis</i> Hendel	b	3	Ghana	22
	<i>Ceratitis quilicci</i> De Meyer	b	3	Francia	24
	<i>Mwatawala & Virgilio</i>				25
	<i>C. cosyra</i> Walker	b	3	Kenia	27
	<i>C. rosae</i> Karsch	b	3	Tanzania	24
	<i>C. capitata</i> Wiedemann	b	3	Tanzania	24
				Venezuela	28
				Brasil	29
Lonchaeidae	<i>Anastrepha distincta</i> Greene	b	3	Brasil	30
	<i>Neosilba pseudozadolicha</i> Strikis	b	3	Brasil	31
	<i>N. glaberrima</i> Wiedemann			Brasil	30
	<i>N. zadolicha</i> McAlpine & Stelyskal	b	3	Brasil	32
	<i>N. batesi</i> Curran	b	3	México	33
Neriidae	<i>Nerius pilifer</i> Fabricius,	b	3	Brasil	16
	<i>Olyphiidops carrerai</i> Aczel	b	3	Brasil	16
Curculionidae	<i>Prionomerus anonicola</i> Bondar	a,b	1-4	Brasil	10
	<i>Compsus argyreus</i> L.	a	1	Brasil	9
	<i>Cratosomus bombina</i> Fabricius	a	4		16
	<i>C. inaequalis</i> Champion	b	4	Venezuela	11
	<i>Optatus palmaris</i> Pascoe	a,b	1,2,3	México	4,34-35
Bostriichidae	<i>Xylosandrus compactus</i> Eichhoff	a,b	4	Brasil	36
	<i>Euwallaceae nr. fornaticus**</i>	a,b	4	Estados Unidos	37
	<i>Apate monacha</i> Fabricius	a,b	4	Puerto Rico	13

Melolonthidae	<i>Euphoria leucographa</i> Gory & Percheron	a	3	México	2-4,38
Chrysomelidae	<i>Omophoita aequinoctialis</i> L.	a	1	Brasil	16
Cerambycidae	<i>Pelurica aemula</i> Weise	a	1	Brasil	16
	<i>Trachyderes mandibularis</i>	a	3	México	39
	Dupont				
	<i>Acanthoderes quadrigibba</i> Say	b	4	México	40
	<i>Oreoderes fasciculosa</i> Thomson	b	4	México	40
	<i>Ateralphus senilis</i> Bates	b	4	Guyana Francesa Brasil Perú Bolivia	41
Buprestidae	<i>Chrysobothris totonaca</i>	b	4	México	40
Coreidae	Domínguez y Márquez				
	<i>Acanthocephala femorata</i>	a,c	3	México	2-3
	Fabricius				
	<i>Acanthocephala alata</i> Burmeister	a,c	3	México	42
	<i>Piezogaster odiosus</i> Stal.	a,c	3	México	42
	<i>Acantocephala parensis</i> Dallas	a,c	1,3	Brasil	16
Coccidae	<i>Coccus hesperidium</i> L.	a,c	3-4	México	4,43
	<i>Parasaissetia nigra</i> Nietner	a,c	3-4	Honduras	7
	<i>Saissetia hemisphaerica</i>	a,c	3-4	Brasil	9
	Targioni-Tozzetti				
	<i>Ceroplastes floridensis</i> Comstock	a,c	1,3-4	Costa Rica	14
	<i>Ceroplastes sinensis</i> Del Guercio	a,c	1,3,4	Costa Rica	14
		a,c	1,3-4	Puerto Rico	5
	<i>Saissetia coffeae</i> Walker	a,c	1,3-4	Puerto Rico	5
	<i>S. oleae</i>	a,c	3	Honduras	7
	<i>S. neglecta</i> De Lotto	a,c	3	Inglaterra***	44
	<i>Coccus longulus</i> Douglas				
	<i>Phalacrococcus howertonii</i>	a,c	4	Estados Unidos	45
	Hodges & Hodgson				
	<i>Hemilecanium guanabana</i> Kondo	a,c	3	Colombia	46
	& Hodgson			Costa Rica	5
	<i>Philephedra broadwayi</i> Cockerell	a,c	1,3-4	Puerto Rico	14
Asterolecaniidae	<i>P. tuberculosa</i> Nakah-Gill	a,c	1,4	Inglaterra***	44
	<i>Asterolecanium pustulans</i>	a,c	1,3-4	Puerto Rico	5
	Cockerell	a,c	1,4	Puerto Rico	5
Pentatomidae	<i>Chlorocoris atrispinus</i> Stal	a,c	3	México	43
	<i>Antiteuchus sepulchralis</i>	a,c	1	Brasil	16
	Fabricius	a,c		Colombia	45
Pseudococcidae	<i>Maconellicoccus hirsutus</i> Green		1-4	Brasil	47-49
				México	4
	<i>Dysmicoccus brevipes</i> Cockerell	a,c	1-4	Inglaterra***	44
	<i>Ferrisia terani</i> Williams &	a,c		México	4
	Granara de Willink				
	<i>F. virgata</i> Cockerell	a,c	1-4	Brasil	49
	<i>F. dayslirii</i> Kaydan & Gullan	a,c	1-4		
	<i>F. malvastra</i> MacDaniel	a,c	1-4		
	<i>Planococcus minor</i> Maskell	a,c	1-4		
	<i>Phenacoccus solenopsis</i> Tinsley	a,c	1-4		
	<i>Pseudococcus jackbeardsleyi</i>	a,c	1-4		
	Gimpel & Miller				
	<i>D. brevipes</i>	a,c	1-4	Inglaterra***	44
	<i>Planococcus citri</i> Risso	a,c	1-4	Puerto Rico	5

				Costa Rica	14
				México	4
				Inglaterra***	44
	<i>P. minor</i>	a,c	3		
	<i>Nipaecoccus nipae</i> Maskell				
	<i>Pseudococcus longispinus</i>	a,c	2,4	Puerto Rico	5
	Targioni-Tozzetti	a,c		Inglaterra***	44
				México	43
	<i>N. nipae</i> Maskell	a,c	1,4	Puerto Rico	5
	<i>P. minor</i>	a,c	1	Estados Unidos	50
Membracidae	<i>Membracis foliata</i> L.	a,c	4	Brasil	9
	<i>M. suctifructus</i> Boulard & Couturier	a,c	2-3	Brasil	16,51
	<i>M. tectigera</i> Olivier				
	<i>Enchenopa albidorsa</i> Fairm.	a,c	1,4	Brasil	16
	<i>Guayaquila tenuicornis</i> Walker				
	<i>Amastris obtegens</i> Fabricius				
	<i>Ceresa peruvensis</i> Temes				
	Lenicov				
	<i>Tunelia longula</i> Burm.				
	<i>Stegaspis fronditia</i> L.				
	<i>Cyphonia clavata</i> Fabricius				
	<i>Bocydium globulare</i> Fabricius				
	<i>Leioscyta spiralis</i> Haviland	a,c	3-4	México	4
	<i>Membracis dorsata</i> Fabricius	a,c	3-4	México	4
	<i>M. mexicana</i> Guérin-Méneville	a,c	3-4	México	3
				Honduras	7
	<i>Calloconophora caliginosa</i>	a,c	1-2,4	Honduras	7
	Walker				
Aleyrodidae	<i>Horioila lineolata</i> Farmaire	a,c	3-4	Venezuela	11
	<i>Vanduzea segmentata</i> Fowler	a,c			
Aphididae	<i>Aleurodicus cocois</i> Curtis	a,c	1	Brasil	9
	<i>Aphis spiraecola</i> Patch	a,c	1	México	4
			1-2	Brasil	16
	<i>A. gossypii</i>	a,c	1-2	Brasil	16
			2-3	México	3
	<i>A. citricola</i>	a,c	1	Puerto Rico	5
	<i>Toxoptera aurantii</i> Boyer de Fonscolombe	a,c	1	Costa Rica	14
Tingidae			1-4	Venezuela	11
				Puerto Rico	5
	<i>Corythucha gossypii</i> Fabricius	a,c	1	Venezuela	11
				Puerto Rico	5
				Honduras	7
				Costa Rica	14
				México	52
Diaspididae	<i>Corythucha ciliata</i> Say	a,c	1	México	53
	<i>Teleonemia moria</i> Stoll.	a,c	1,3	Brasil	16
	<i>Pinnaspis strachani</i> Cooley	a,c	3-4	Puerto Rico	5
				Costa Rica	14
				Inglaterra***	44
	<i>P. aspidistrae</i> Signoret	a,c	3-4	Brasil	16
	<i>Clavaspis herculeana</i> Cockerell & Hadden	a	3	Inglaterra***	44
	<i>Aonidiella aurantii</i> Maskell	a,c	1,4	Puerto Rico	5
	<i>A. comperei</i> Comstock				
	<i>Chrysomphalus adonidium</i> L.				
	<i>Mycelaspis personata</i> Comstock				
	<i>Selenaspis articulatus</i> Morgan				
Aethalionidae	<i>Aethaliom reticulatum</i> L.	a,c	2-3	Venezuela	11
Cicadellidae	<i>Empoasca insularis</i> Oman	a,c	1	Cuba	54
Miridae	<i>Hyaloides nani</i> Maldonado	a,c	1	Puerto Rico	5

Monophlebidae	<i>Crypticerya multicicatrices</i> Kondo & Unruh	a,c	1,4	Colombia	55
Termitidae	<i>Nasutitermes corniger</i> Motschulsky	a	4	Venezuela	11
	<i>N. costalis</i> Holmgren	a	4	Puerto Rico	5
Tettigoniidae	<i>Idiarthron subquadratum</i> Saussure & Pictet	a,c	1-3	México	56
Eriophyidae	<i>Aceria annonae</i> Keifer	a,c	1	México	57
		a,c	1	Estados Unidos	58
Tetranychidae	<i>Tetranychus mexicanus</i> McGregor	a,c	1	Brasil	59
		a,c	1	México	60

*Los números de referencia se encuentran en la literatura citada entre paréntesis después del año de publicación. **Se sabe que no es fitófago *per se*. ***Muestras recolectadas en frutos de importación. Estado Biológico: a = adulto, b = larva, y c ninfa. Daños: 1 = hoja, 2 = flor, 3 = fruto, y 4 = rama.

de la región tropical y subtropical. Se determinaron siete órdenes de insectos, 33 familias y uno de ácaros con dos familias (Fig. 1). El orden de insectos con mayor número de especies asociadas con *A. muricata* es Hemiptera con 14 familias que incluyen 70 especies. Dentro de éstas, las más representativas son Membracidae con 17 especies, Coccidae y Pseudococcidae ambas con 13 especies. Le sigue el orden Coleoptera con 16 especies y seis familias, siendo las más, representativas Curculionidae y Cerambycidae con siete y cuatro especies. En tercer lugar, Diptera con 13 especies en tres familias, de las cuales Tephritidae incluye siete especies. Lepidoptera con 10 especies, seis familias; Hymenoptera dos familias: Eurytomidae y Apidae con cuatro y una especie. Dos especies de termitas (Blattodea) de la familia

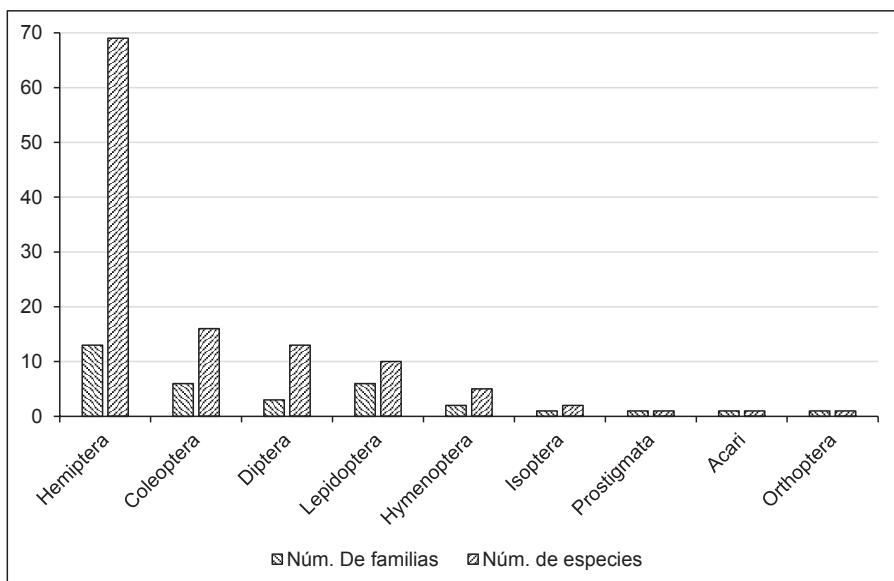


Fig. 1. Número de familias y especies de insectos y ácaros agrupados por orden.
Fig. 1. Number of families and species of insects and mites grouped by order.

Termitidae. Dos especies de ácaros de los órdenes Prostigmata, familias Eriophyidae y Tetranychidae. Orthoptera con una especie de la familia Tettigoniidae.

La mayor cantidad de insectos tienen hábitos barrenadores, masticadores, y chupadores que dañan frutos de *A. muricata* en comparación con otras estructuras de la planta (Cuadro 1). De éstos, los de mayor importancia con base en el número de registros, distribución, daños infringidos al fruto y su asociación con patógenos se ubican los barrenadores de frutos *Bephratelloides cubensis* Ashmead, *B. limae* Bondar, *B. maculicollis* Cameron, *B. pomorum* Fabricius (Hymenoptera: Eurytomidae) (McComie 1987, Medina-Gaud et al. 1989, Granadino y Cave 1997, Hamada et al. 1998, Cabrera y Martínez 2001, Hernández-Fuentes et al. 2008). Estas avispas son de origen Neotropical y se le asocia con siete especies frutícolas del género *Annona* (Grissell y Schauff 1990). En México, se ha observado hasta 60% de infestación en frutos y pérdidas en la producción de 25% causadas por *B. cubensis* (Hernández-Fuentes et al. 2010).

Le sigue en importancia *Cerconota anonella* Sepp. (Lepidoptera: Depressariidae), distribuida principalmente en Centro y Sudamérica. En México únicamente se ha reportado en Veracruz (Ruiz y Flores 2011, Ruiz-Montiel et al. 2014), aunque también se conoce su presencia en frutos de guanábana de traspaso en los estados de Tabasco y Chiapas (comunicación personal). En Brasil se presenta una amplia distribución (Caloba y da Silva 1995, Hamada et al. 1998, Braga et al. 1999, Broglio-Micheletti y Berti-Filho 2000, Da Silva et al. 2006). Braga et al. (1999) mencionan a *C. anonella* como la principal plaga del fruto en el norte de Brasil, puede infestar hasta 73% de los frutos y causar pérdidas en el rendimiento del 50% si no se realiza control (Bustillo y Peña 1992, Broglio-Micheletti y Berti-Filho 2000). La importancia de *C. anonella* es que una sola larva puede provocar que el fruto ya no sea comercializado, ya sea por el daño directo al alimentarse, o bien porque permite el libre acceso de insectos y patógenos oportunistas que contribuyen a la pudrición del fruto.

De las 119 especies fitófagas asociadas con *A. muricata*, se encontró que las partes vegetales más afectadas son el fruto, las ramas y las hojas con 67 (56.8%), 60 (50.8%), y 57(48.3%) especies, respectivamente (Fig. 2). Mientras que para las flores solo se identificaron 23 (19.5%) especies, en tanto que el tronco fue el menos afectado con solo 3 especies (2.5%).

El estado biológico predominante que se encontró causando daño fue el adulto con 88 reportes que equivalen al 74.6%, seguido del estadio ninfal con 67 especies representando el 56.8% y en etapa larvaria en menor número (39) siendo 33.1% (Fig. 3). Los esfuerzos para el control de plagas y otros estudios podrían dirigirse a los adultos y ninfas principalmente.

Las plagas se pueden encontrar en más de un estado biológico dañando a *A. muricata* (Fig. 4). Se detectó que aproximadamente la mitad afectan solo una parte vegetal, y el resto dos o más parte vegetales. Dos terceras partes de las plagas se reportaron con 2 estadios biológicos diferentes en *A. muricata*.

Mediante revisión de literatura, colecciones de insectos y observaciones de campo Peña y Bennet (1995) elaboraron una lista reportaron 48 especies de insectos como plagas asociadas con *A. muricata*, no obstante, algunos reportes son dudosos puesto que los especímenes fueron recolectados en estado adulto sin verificar la condición fitófaga en el hospedante, y por la posibilidad de utilizar la planta como refugio, sitio de apareamiento o fuente de néctar o azúcares de frutos maduros o en descomposición. Como ejemplo de lo anterior se menciona la relación de *Drosophila melanogaster* Meigen, *D. cardinoides* Dobzhansky & Pavan en *A. muricata*, en donde

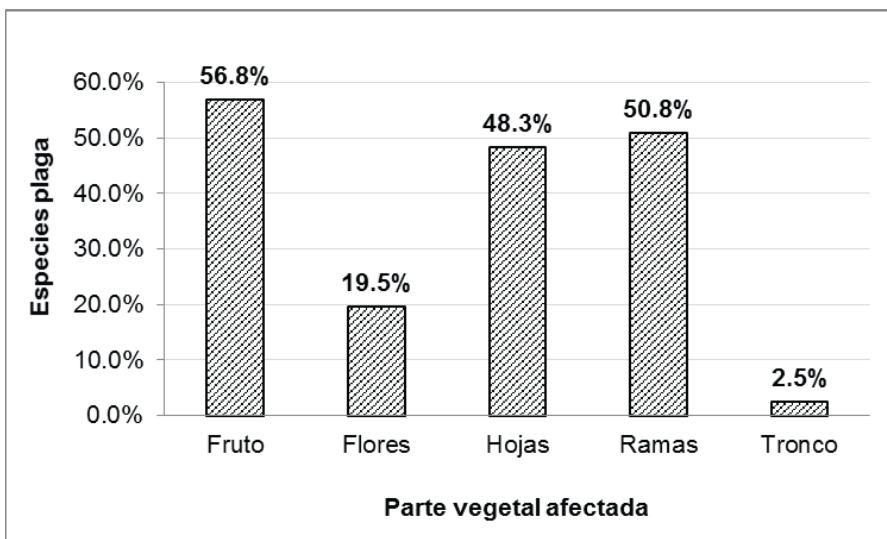


Fig. 2. Porcentaje de especies plaga por estructura vegetal en *Annona muricata*.
Fig. 2. Percentage of pest species in each *Annona Muricata* plant structure.

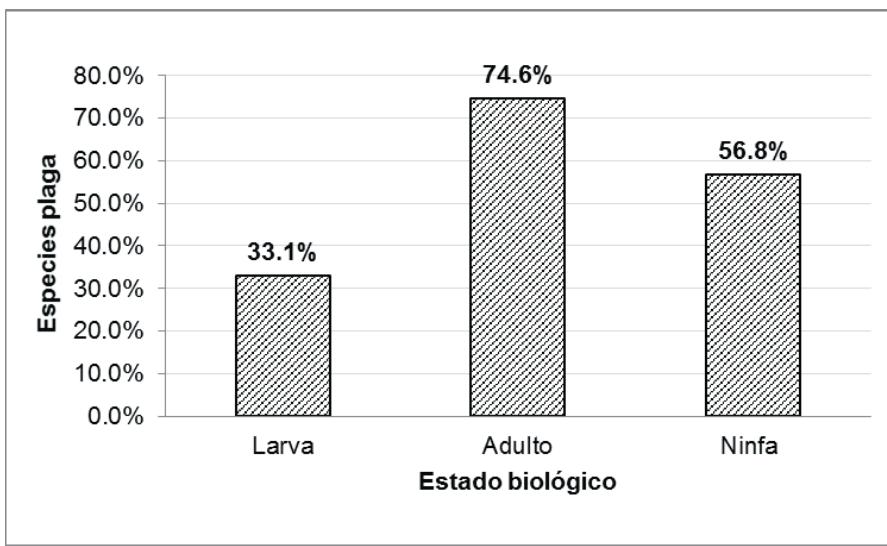


Fig. 3. Porcentaje de especies plaga de acuerdo con el estado biológico que causa daño en *Annona muricata*.
Fig. 3. Percentage of pest species according to the biological state that causes damage in *Annona muricata*.

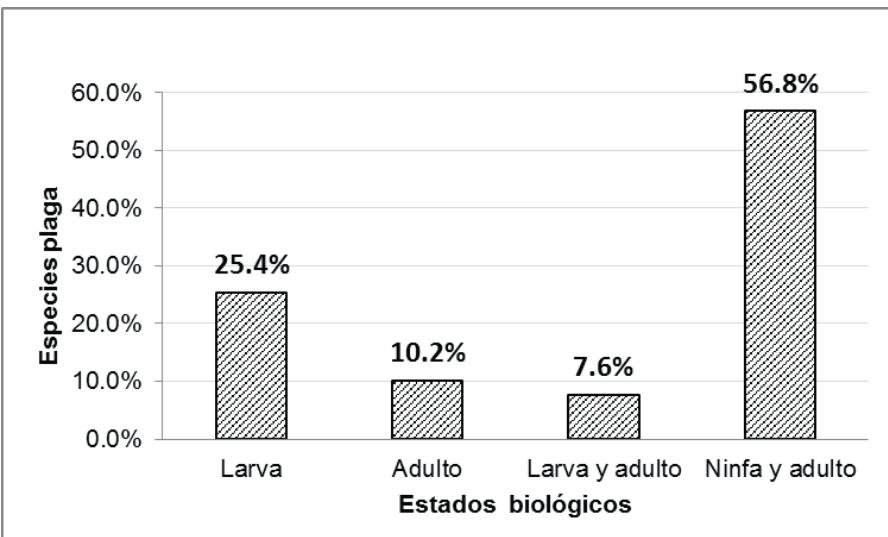


Fig. 4. Porcentaje de especies plaga que afectan *Annona muricata* en sus diferentes estados de desarrollo.

Fig. 4. Percentage of pest species that affect *Annona muricata* in their different biological stages.

el insecto se alimenta tanto en larva como adulto de frutos maduros adheridos al árbol o caídos en proceso de descomposición sin ser necesariamente una plaga (Bedichek 1965). Así mismo las hembras de *Deciroprocta bulgara* (Distant) ovipositan en varias especies vegetales incluida *A. muricata*, en la cual prefieren las ramas de los árboles o plantas jóvenes como sitios de oviposición (Sánchez-García et al. 2019). De la misma manera se ha observado que el adulto de *Cyclocephala brittoni* en *A. muricata* (Ratcliffe 1992) y en inflorescencias de otras especies de plantas cuya función es la polinización (Schatz 1990). *Bephratelloides macullicolis*, *B. pomorum*, y *B. limai* se encuentran en Brasil, consideradas como plagas de mayor importancia en la guanábana (Caloba y da Silva 1995, Hamada et al. 1998, Braga et al. 1999, Santos et al. 2001). En México sólo se ha reportado la presencia de *B. cubensis* (Hernández-Fuentes et al. 2010, Ruiz-Montiel y Flores 2011, Cham et al. 2019). *A. quadrigibba*, *O. fasciculosa*, y *C. totonaca* son insectos oportunistas que infestan al guanábano cuando éste se encuentra dañado o debilitado por enfermedades; no obstante, de acuerdo a Hernández-Fuentes et al. (2018) la infestación puede acentuar el daño por patógenos. *Optatus palmaris* con mayor presencia y distribución en México se ha vuelto una plaga de importancia en los últimos años en el cultivo de guanábana ya que llega a infestar hasta 79.3% de los frutos con una severidad en éstos del 38 al 98% (Maldonado-Morales et al. 2014), para su control se han evaluado distintos productos insecticidas de origen químico y extractos vegetales con resultados satisfactorios (Hernandez-Fuentes et al. 2021), asimismo se ha determinado la feromona sexual de agregación α -terpineol con el objetivo de utilizar técnicas de menor impacto ambiental para su manejo (Pineda-Ríos et al. 2021).

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Referencias Citadas

- Adaime, R., M. S. Miranda de S., C. R. de Jesus B., E. da Glória de D., J. F. Pereira, P. C. Strikis, and M. F. de Souza F. 2017 (31). Frugivorous flies (Diptera: Tephritidae, Lonchaeidae), their host plants, and associated parasitoids in the extreme north of Amapá State, Brazil. *Fla. Entomol.* 2: 316-324.
- Aluja, M., J. Guillén, G. de la Rosa, M. Cabrera, H. Celedonio, P. Liedo, and J. Hendrichs. 1987. Natural host plant survey of the economically important fruit flies (Diptera: Tephritidae) of Chiapas, Mexico. *Fla. Entomol.* 3: 329-338.
- Badii, K. B., M. K. Billah, K. Afreh-Nuamah, and D. Obeng-Ofori. 2015. (25). Species composition and host range of fruit-infesting flies (Diptera: Tephritidae) in northern Ghana. *Int. J. Trop. Insect Sci.* 3: 137-151. <http://journals.cambridge.org/action/>.
- Bedichek, P. S. 1965. The influence of adult and larval food habits on population size of neotropical ground-feeding *Drosophila*. *Am. Midl. Nat.* 1: 1-27. <http://www.gorgas.gob.pa/BiblioGorgas/pdf/The%20Influence%20of%20Adult%20and%20Larval%20Food%20Habits%20on%20Population%20Size%20of%20Neotropical%20Ground%20Feeding%20Drosophila%20by%20Sarah%20Bedichek%20Pipkin.pdf>.
- Braby, M. F., D. C. Franklin, D. E. Bisa, M. R. Williams, A. A. E. Williams, C. L. Bishop, and R. A. M. Coppin. 2021. (22). *Atlas of Butterflies and Diurnal Moths in the Monsoon Tropics of Northern Australia*. ANU Press, Canberra, Australia. <http://doi.org/10.22459/ABDM.12.2018>.
- Braga, S. R., C. T. Bandeira, and A. L. M. Mesquita. 1999. (10). Occurrence and damage of soursop pests in northeast Brazil. *J. Crop Prot.* 18: 539-541.
- Broglia-Micheletti, S. F., y E. Berti-Filho. 2000. (17). Controle de *Cerconota anonella* em pomar de gravoleira. *Sci. Agric.* 3: 557-559. <https://doi.org/10.1590/S0103-90162000000300029>.
- Brunner, S.C., y J. Acuna. 1967. (8). Sobre la biología de *Bephrrata cubensis* Ashm., el insecto perforador de las frutas anonáceas. Academia Ciencias Cuba. Inst. Agron. Ser. Agr. 1: 14.
- Bustillo, A. E., and J. E. Peña. 1992. (19). Biology and control of the *Annona* fruit borer *Cerconota anonella* (Lepidoptera: Oecophoridae). *Fruits* 1: 81-84.
- Cabrera, I., y S. Martínez. 2001. (13). Susceptibilidad a insectos en selecciones y variedades de *Annona muricata* L. y *Persea americana* M. en Puerto Rico. *Agron. Mesoam.* 1: 99-103. <https://www.redalyc.org/articulo.oa?id=43712114>
- Caloba, J., e N. M. da Silva. 1995. (9). Insetos associados a graviola, *A. muricata* L. e biribá, *Rollinia mucosa* Jacq. *Bail no estado do Amazonas. Ann. Soc. Entomol. Brasil* 1: 179-182. <https://doi.org/10.1590/1809-43921998284431>.
- Cambero, N. K. G., O. J. Cambero C., M. Rodríguez P., A. Robles B., N. Isiordia A., C. B. Cambero A., y G. S. Bayardo C. 2021. (52). Primer registro de *Corythucha gossypii* en *Annona muricata* L. en México. *Southwest. Entomol.* 2: 557-562.

- Cambero-Ayón, C. B., M. Rodríguez-Palomera, J. C. Cambero-Campos, A. K. Cham, y K. G. Cambero-Nava. 2017. (38). *Euphoria leucographa* (Gory & Percheron, 1833) (Coleoptera: Melolonthidae) en frutos de guanábana (*Annona muricata* L.) en Nayarit, México. Rev. Gadit. Entomol. 1: 223-227.
- Carrillo, D. J. E. Peña, J. H. Crane, and R. E. Duncan. 2006. (50). Pollinator and Insect/Mite Management in *Annona* spp. ENY-834. UF/IFAS Extension. <https://edis.ifas.ufl.edu/publication/IG166>
- Cham, A. K., G. Luna-Esquivel, A. Robles-Bermúdez, C. Ríos-Velasco, J. A. Coronado-Blanco, and O. J. Cambero-Campos. 2019. (4). Insects associated with the soursop (*Annona muricata* L.) crop in Nayarit, Mexico. Fla. Entomol. 2: 359-365.
- Copeland, R., R. A. Wharton, Q. Luke, M. De Meyer, S. Lux, N. Zenz, P. Machera, and M. Okumu. 2006. (27). Geographic distribution, host fruit, and parasitoids of African fruit fly pests *Ceratitis anoneae*, *Ceratitis cosyra*, *Ceratitis fasciventris*, and *Ceratitis rosa* (Diptera: Tephritidae) in Kenya. Ann. Entomol. Soc. Am. 2: 261-278.
- Coto, A. D., y J. L. Saunders. 2001. (14). Insectos plaga de la guanábana (*Annona muricata*) en Costa Rica. Manejo Integrado de Plagas 61: 60-68. <https://repositorio.catie.ac.cr/handle/11554/6416>.
- Couturier, G. 1992. (51). Conocimiento y manejo de los insectos plagas de los frutales de la Amazonía. Folia Amazónica 1: 31-39. <https://doi.org/10.24841/fa.v4i1.176>.
- Da Silva, E. L., C. M. de Carvalho, R. R. do Nascimento, A. L. Mendonca, C. E. da Silva, G. B. Goncalves, M. R. T. de Freitas, and A. E. G. Sant'Ana. 2006. (18). Reproductive behaviour of the *Annona* fruit borer, *Cerconota anonella*. Ethology 112: 971-976. <https://doi.org/10.1111/j.1439-0310.2006.01255.x>.
- De Sousa, J. M., M. G. C. Gondim, and A. Lofego. 2010. (58). Biología de *Tetranychus mexicanus* (McGregor) (Acari: Tetranychidae) em três espécies de Annonaceae. Neotrop. Entomol. 3: 319-323.
- Do Nascimento, L. L., R. Adaime, S. V. Costa N., E. G. de Deus, C. R. Jesus B., and P. C. Strikis. 2015. (32). New findings on Lonchaeidae (Diptera: Tephritoidea) in the Brazilian Amazon. Fla. Entomol. 4: 1227-1237.
- Do Nascimento, L. L., E. G. de Deus, D. B. do Nascimento, C. R. Jesus B., S. V. da Costa N., and R. Adaime. 2017. (30). Species of *Anastrepha* (Diptera: Tephritidae), their host plants, and parasitoids in small fruit production areas in the State of Amapá, Brazil. Fla. Entomol. 2: 403-410.
- Domínguez, G. O. E. 1978. (11). Insectos perjudiciales del guanábano (*Annona muricata* L.) en el estado de Zulia, Venezuela. Revista de la Facultad de Agronomía 3: 149-163. <https://produccioncientificafaluz.org/index.php/agronomia/article/view/25815>.
- Elton, C. 1927. Animal Ecology. Macmillan Company, New York.
- Flores-Canales, R. J., J. A. Acuña-Soto, C. Santillán-Ortega, N. Işıordia-Aquino, A. M. Sotelo-Montoya, y R. D. Hernández-Zaragoza. 2019. (57). Fluctuación poblacional de *Aceria annoneae* (Keifer, 1973) (Prostigmata: Eriophyidae), en tres municipios de Nayarit, México. Revista Mexicana de Ciencias Agrícolas 1: 177-186. <https://doi.org/10.29312/remexca.v10i1.1734>.
- Forti, B. S. M., E. Prado C., J. M. Dos Santos, y L. Broglia M. 2015. (47). Registro da cochinilha-rosada-do-hibisco infestando frutíferas em Maceió, Alagoas, Brasil. Revista Caatinga 2: 242-248. <https://periodicos.ufersa.edu.br/caatinga/article/view/3756>.

- Granadino, A. C., and R. D. Cave. 1997. (7). Within-tree distribution of seven insect pests of soursop (*Annona muricata*) in Honduras. *Ceiba* 2: 161-166. <https://revistas.zamorano.edu/index.php/CEIBA/article/view/159>.
- Grissell, E. E., and M. E. Schauf. 1990. A synopsis of the seed-feeding genus *Bephratelloides* (Chalcidoidea: Eurytomidae). *Proc. Entomol. Soc. Wash.* 2: 177-187.
- Hamada, N., A. L. S. Gomez, G. Couturier, y B. Ronchi-Teles. 1998. (16). Insetos associados a gravioleira (*Annona muricata* L., Annonaceae) na regiao de Manaus, Amazonas, Brasil. *Acta Amaz.* 4: 425-431. <https://doi.org/10.1590/1809-43921998284431>.
- Hernández-Fuentes, L. M., y J. M. Pinzón-García. 2015. (42). Primer reporte de *Piezogaster odiosus* Stal y *Acanthocephala alata* Burmeister (Hemiptera: Coreidae) como plagas de *Annona muricata* L. *Southwest. Entomol.* 3: 665-668.
- Hernández-Fuentes, L. M., M. A. Urias-López., y N. Bautista-Martínez. 2010. (1). Biología y hábitos del barrenador de la semilla *Bephratelloides cubensis* Ashmead (Hymenoptera: Eurytomidae). *Neotrop. Entomol.* 4: 527-534. <https://doi.org/10.1590/S1519-566X2010000400010>.
- Hernández-Fuentes, L. M., J. M. Valdés-Carrasco, M. A. Urías-López, y R. Gómez-Jaimes. 2015. (20). Identificación, daños y fluctuación poblacional de *Gonodonta pyrgo* (Lepidoptera: Noctuidae) en *Annona muricata*. *Revista Mexicana de Ciencias Agrícolas* 5: 1001-1012. <https://doi.org/10.29312/remexca.v6i5.594>.
- Hernández-Fuentes, L. M., A. Castañeda-Vildózola, M. Orozco-Santos, y J. Valdés-Carrasco. 2016. (56). El chacuatete, *Idiarthron subquadratum* en guanábana, *Annona muricata*. *Southwest. Entomol.* 3: 855-864.
- Hernández-Fuentes, L. M., R. Gómez-Jaimes, V. López-Martínez, y A. Castañeda-Vildozola. 2018. (40). Insectos xilófagos asociados a ramas de guanábano (*Annona muricata* L.) afectados por *Lasiodiplodia theobromae* Pat. *Southwest. Entomol.* 2: 543-546.
- Hernández-Fuentes, L. M., J. M. Pineda-Ríos, y M. Orozco-Santos. 2020. (39). Primer reporte de *Trachyderes (Dendrobias) mandibularis* Dupont asociado a guanábana (*Annona muricata* L.). *Southwest. Entomol.* 1: 325-328.
- Hernández-Fuentes, L. M., Y. Nolasco-González, M. Orozco-Santos, y E. Montalvo-González. 2021. (35). Toxicidad de insecticidas contra *Optatus palmaris* Pascoe en guanábana. *Revista Mexicana de Ciencias Agrícolas* 1: 49-60. <https://doi.org/10.29312/remexca.v12i1.2417>.
- Hodges S. G., and C. J. Hodgson. 2010. (45). *Phalacrocooccus howertoni*, a new genus and species of soft scale (Hemiptera: Coccidae) from Florida. *Fla. Entomol.* 1: 8-23.
- Illescas-Riquelme, C. P., H. González-Hernández, J. Valdés-Carrasco, M. C. M. Llanderol-Cázares, and C. Ruiz-Montiel. 2015. (33). Lonchaeidae (Diptera: Tephritoidea) associated with the genus *Annona* in Mexico. *Southwest. Entomol.* 1: 121-130.
- Kondo, T., and C. Hodgson. 2013. (46). A third species of *Hemilecanium* Newstead (Hemiptera: Coccoidea: Coccidae) from the New World, with keys to species in the genus. *Neotrop. Entomol.* 5: 508-520.
- Kondo, T., and C. M. Unruh. 2009. (55). A new species of *Crypticerya* Cockerell (Hemiptera: Monophlebidae) from Colombia, with a key to species of the tribe Iceryini found in South America. *Neotrop. Entomol.* 38: 92-100.

- Laca, M. d L., y L. Ramón. 1999. Las plantas americanas en la obra de Charles de L'Écluse: primeras citas en las cartas de Juan Castañeda. *Anales Jard. Bot.* Madrid 57: 97-107. DOI: 10.3989/ajbm.1999.v57.i1.192.
- Lemos, P. d. E. E. 2011. Panorama de las anonas cultivadas en Brasil: Saramuyo, guanábana y atemoya, pp. 21-36. In E. A. R. González, C. L. M. Luna, J. J. Gutiérrez, G. M. A. Schlie, y L. D. G. Vidal [eds.], Anonáceas: Plantas Antiguas, Estudios Recientes; Colección Jaguar Unicach: Chiapas, México. ISBN: 978-607-7510-91-8.
- Maldonado-Jiménez, E., L. M. Hernández-Fuentes, G. Luna Esquivel, R. Gómez-Jaimes, R. J. FloresCanales, y M. Orozco-Santos. 2014. (34). Bioecología de *Optatus palmaris* Pascoe (Coleoptera: Curculionidae) en *Annona muricata* L. Southwest. *Entomol.* 4: 773-782.
- Malumphy, C. 2017. (44). Scale insects (Hemiptera: Coccoidea) intercepted in England on fresh soursop, *Annona muricata*, fruit imported from the Neotropical region. *Entomologist's Gazette* 4: 263-271.
- Martins, D. S., M. J. Fornazier, A. L. B. G. Peronti, M. P. Culik, C. A. S. Souza, R. C. Taques, J. S. Znuncio J., and R. B. Queiroz. 2019. (48). *Maconellicoccus hirsutus* (Hemiptera: Pseudococcidae) in Brazil: recent spread, natural enemies, and new hosts. *Fla. Entomol.* 2: 438-443.
- McComie, L. D. 1987. (12). The soursop (*Annona muricata* L.) in Trinidad: its importance, pests and problems associated with pest control. *J. Agric. Soc. Trinidad and Tobago* 87: 42-55. <https://www.cabi.org/isc/abstract/19901122378>.
- Medina-Gaud, S., Bennett, F. D., Segarra-Carmona, A. E., and Pantoja, A. 1989. (5). Notes on insect pests of soursop (guanabana), *Annona muricata* L. and their natural enemies in Puerto Rico. *Journal of Agriculture of the University of Puerto Rico* 4: 383-389. <https://doi.org/10.46429/jaupr.v73i4.6318>.
- Monné, M. A., and C. S. Chaboo. 2015. (41). Beetles (Coleoptera) of Peru: a survey of the families Cerambycidae, Disteniidae, and Vesperidae. *J. Kans. Entomol. Soc.* 1: 34-120.
- Moquet, L., J. Payet, S. Glenac, and H. Delatte. 2021. (26). Niche shift of tephritid species after the Oriental fruit fly (*Bactrocera dorsalis*) invasion in La Réunion. *Divers. Distrib.* 1: 109-129. <https://doi.org/10.1111/ddi.13172>.
- Morales, P., M. Cermeli, F. Gody, y B. Salas. 2004. (28). Lista de hospederos de la mosca del Mediterráneo *Ceratitis capitata* Wiedemann (Diptera: Tephritidae) basada en los registros del Museo de Insectos de Interés Agrícola del INIA – CENIAP. *Entomotropica* 1: 51-54. http://saber.ucv.ve/ojs/index.php/rev_ent/article/view/7567.
- Mwatawala, M. W., M. De Meyer, R. H. Makundi, and A. P. Maerere. 2009. (24). Host range and distribution of fruit-infesting pestiferous fruit flies (Diptera, Tephritidae) in selected areas of Central Tanzania. *Bull. Entomol. Res.* 99: 629-641.
- Nadel, H., and J. Peña. 1991. (6). Hosts of *Bephratelloides cubensis* (Hymenoptera: Eurytomidae) in Florida. *Fla. Entomol.* 3: 476-479.
- Oliveira, M. C., C. A. H. Flechtmann, and M. R. Frizzas. 2008. (36). New findings on Lonchaeidae (Diptera: Tephritoidea) in the Brazilian Amazon. *Coleopt. Bull.* 1: 45-48.
- Owens, D., L. F. Cruz, W. S. Montgomery, T. I. Narvaez, E. Q. Schnell, N. Tabanca, R. E. Duncan, D. Carrillo, and P. E. Kendra. 2018. (37). Host range expansion

- and increasing damage potential of *Euwallacea* nr. *fornicatus* (Coleoptera: Curculionidae) in Florida. Fla. Entomol. 2: 229-236.
- Pacheco, S. V. C., M. Bora K., C. S. S. Da Silva T., and J. Braz T. 2019. (49). Mealybug species (Hemiptera: Coccoidea: Pseudococcidae) on soursop and sugar apple (Annonaceae) in North-East Brazil, with description of a new species of *Pseudococcus* Westwood. Zootaxa 3: 525-538.
- Palemón-Alberto, F., S. A. Ortega-Acosta, A. Castañeda-Vildózola, S. Domínguez-Monge, E. Cruz-Crespo, C. R. Juárez-Rosete, G. Reyes-García. 2021. (53). New record of *Corythucha ciliata* (Say, 1932) damaging *Annona muricata* L. at Guerrero and Puebla, Mexico. Southwest. Entomol. 2: 553-556.
- Peña, J. E., and F. D. Bennett. 1995. Arthropods associated with *Annona* spp. in the Neotropics. Fla. Entomol. 2: 329-349.
- Pineda-Ríos, J. M., J. Cibrian-Tovar, L. M. Hernández-Fuentes, R. M. López-Romero, L. Soto-Rojas, J. Romero-Nápoles, C. Llanderal-Cázares, L. F. Salomé-Abarca. 2021. α-Terpineol: an aggregation pheromone in *Optatus palmaris* (Coleoptera: Curculionidae) (Pascoe, 1889) enhanced by its host-plant volatiles. Molecules 10:1-15. <https://doi.org/10.3390/molecules26102861>.
- Pinzón-García, J. M., L. M. Hernández-Fuentes, G. Luna-Esquível, N. Isiordia-Aquino, y M. Ortíz-Catón. 2016. (21). Biología y hábitos de *Gonodonta pyrgo* Cramer en *Annona muricata*. Southwest. Entomol. 1: 251-258.
- Prasad, M., H. Singh, and S. K. Singh. 2014. Early Eocene *Annona* fossils from Vastan Lignite Mine, Surat District, Gujarat, India: age, origin and palaeogeographic significance. Curr. Sci. 107: 1730-1735. <https://www.jstor.org/stable/24107949>.
- Raga, A., M. F. de Souza F., R. A. Machado, M. E. Sato, and R. C. Siloto. 2011. (29). Host ranges and infestation indices of fruit flies (Tephritidae) and lance flies (Lonchaeidae) in São Paulo State, Brazil. Fla. Entomol. 4: 787-794.
- Ratcliffe, C. B. 1992. Nine new species and 11 country records of *Cyclocephala* (Coleoptera: Scarabaeidae: Dynastinae) from Panama and Costa Rica. Coleopt. Bull. 3: 216-235.
- Ruiz-Montiel, C., C. P. Illescas-Riquelme, R. Flores-Peredo, L. Vidal-Hernández, y P. I. Domínguez-Espinoza. 2011. (2). Nuevo reporte de incidencia de *Acantocephala femorata* F. (Hemíptera: Coreidae) y *Euphorbia leucographa* G. & P. (Coleoptera: Melolonthidae) en frutos de *Annona muricata* L. en Veracruz, México, pp. 449-456. In A. R. González E., L. M. Luna C., J. Gutiérrez J., M. A. Schlie G., y D. G. Vidal L. [coords.], Anonáceas: Plantas Antiguas, Estudios Recientes. UNICACH, Chiapas, México. ISBN: 978-607-7510-91-8.
- Ruiz-Montiel, C., P. I. Domínguez-Espinoza, R. Flores-Peredo, y C. P. Illescas-Riquelme. 2014. (3). Insects associated with soursop (*Annona muricata* L.) en Veracruz, México. Southwest. Entomol. 2: 367-374.
- Ruiz-Montiel, M. C., N. Reyes P., M. Abato Z., N. Domínguez G., y C. Anastacio L. 2020. (59). *Tetranychus mexicanus* asociado a guanábano (*Annona muricata*) en Veracruz, México. Southwest. Entomol. 3: 815-818.
- San Martín, R. E., P. Ceballos H., A. Salinas C., y C. Ruiz M. 2015. (43). Identificación de Hemípteros asociados al guanábano (*Annona muricata* L.) en Veracruz, México, pp. 145-159. In E. Vidal L., N. A. Vidal M., y L. Vidal H. [comps.] Anonáceas: Plantas Antiguas, Estudios Recientes. Parte 2. Universidad Autónoma Chapingo, Edo. de México, México. ISBN: 978-607-12-0411-0.

- Sánchez-Castro, A., y B. M. Fernández-Argundín. 2018. (54). Informe de *Empoasca insularis* Oman en guanábana (*Annona muricata L.*) en Cuba. Rev. Protección Veg. 1: 1-8. <http://ref.scielo.org/6vcyyd>.
- Sánchez-García, J. A., J. J. Velázquez-Monreal, H. M. Guzmán-Vásquez, R. Jarquín-López, and J. A. Ortiz-López. 2019. New host records and biological notes for *Diceroprocta bulgara* (Distant) in Mexico. Southwest. Entomol. 1: 229-233.
- Santos, O. M. A., N. T. Vilela J., R. Teixeira A., I. Midori I., J. N. Sombra O., and G. Alves A. 2001. (15). Broca da semente da graviola no Distrito Federal. Comunicado Técnico 54. Embrapa, Brasil.
- Schatz, A. B. 1990. Some aspects of pollination biology in Central American forests, pp. 69-84. In Reproductive ecology of tropical forest plants. K. S. Bawa and M. Hadley [eds.], Unesco and the Parthenon Publishing Group, Paris, France. ISBN: 0929858220.
- SIAP (Servicio de Información Agroalimentaria y Pesquera). 2019. <https://nube.siap.gob.mx/cierreagricola/> Consulta 16 Septiembre.
- Vu, T. N., R. Eastwood, C. T. Nguyen, and L. V. Pham. 2008. (23). *Graphium agamemnon* Linnaeus (Lepidoptera: Papilionidae), a pest of soursop (*Annona muricata* Linnaeus), in Vietnam: biology and a novel method of control 38: 174-177. <https://doi.org/10.1111/j.1748-5967.2008.00153.x>.

Lagochile occidentalis*¹ una Nueva Especies para Jalisco, México**Lagochile occidentalis*¹ a New Species for Jalisco, Mexico**

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Resumen. Se describe una nueva especie de Coleóptera: Rutelinae, *Lagochile occidentalis*, capturada con carpotrampas aéreas de una zona de ecotono con elementos de bosque de encino, bosque de pino, bosque mesófilo, de montaña y bosque tropical subcaducifolio en Jalisco. Se proveen ilustraciones de los caracteres distintivos de las especies del género presentes en México, así como de la variación cromática dorsal.

Abstract. A new species of Coleoptera: Rutelinae beetle, *Lagochile occidentalis*, collected with aerial fruit traps from an ecotone zone with oak, pine, tropical subdeciduous, and cloud forest in Jalisco is described. Diagnostic characters are illustrated for the species, and its dorsal chromatic variation.

Introducción

El género *Lagochile* Hoffmannsegg, 1817 (= *Chasmodia* MacLeay, 1819) está constituido por 64 especies americanas cuya distribución abarca desde México hasta Argentina (Soula 2005). Poco se conoce sobre su biología y, Jameson y Morón (2001) describieron la larva de *L. collaris* Blanchard, 1850 como *Chasmodia collaris*, mientras que Albertoni et al. (2014) describieron la larva y la pupa de *L. emarginata* (Gyllenhal, 1817). El mayor número de especies se conoce en Sudamérica (Soula 2005).

Las especies mexicanas de *Lagochile* previamente se ubicaron en el género *Chasmodia* (Delgado 1997, Morón 1997, Jameson y Morón 2001). Desde el reporte de Blanchard (1850) y Soula (2005), se ha establecido que el género válido debe ser *Lagochile*. Para México se conocen dos especies: *L. collaris* Blanchard, 1850: 204 y *L. jamensonae* (Delgado 1997: 17). *Lagochile collaris* ha sido registrada en México en las vertientes del Golfo y del Atlántico, en localidades de Veracruz, Tabasco, Oaxaca, Chiapas (Delgado 1997), y Yucatán (Reyes Novelo y Morón 2005). Además, ha sido registrada en Nicaragua, Costa Rica y Panamá (Morón 1997). Se considera que *Lagochile jamensonae* es endémica de México, conocida en la vertiente del Pacífico, exclusivamente del estado de Guerrero (Delgado 1997).

Como parte del trabajo de campo que se ha realizado en el Jardín Haraveri, Municipio de San Sebastián del Oeste, Jalisco, se han colectado adultos de

¹Scarabaeidae: Rutelinae: Rutelini

Coleoptera de varias familias, utilizando varios métodos de muestreo, entre ellos, colecta directa en hongos, troncos, flores, y otros sustratos, cernido de hojarasca, con necrotrampas y carpotrampas. Con carpotrampas se colectaron varios especímenes adultos pertenecientes al género *Lagochile*. Una revisión detallada de los ejemplares nos permite argumentar que pertenecen a una especie nueva, misma que se describe en este trabajo.

Materiales y Métodos

De julio a noviembre de 2019, se colocaron 20 carpotrampas cebadas con plátano en el Jardín Haraveri, Hacienda Las Tres Carmelitas, San Sebastián del Oeste, Jalisco. Se colocaron cinco carpotrampas por tipo de vegetación: bosque de pino, bosque de pino encino, bosque mesófilo de montaña, y bosque tropical subcaducifolio. A los machos se les extrajo el genital para su revisión bajo un microscopio estereoscópico.

El holotipo y 25 paratipos se encuentran depositados en la Colección Entomológica del Centro de Estudios en Zoología, Universidad de Guadalajara (CZUG); 2 paratipos se encuentran depositados en la Colección Nacional de Insectos, Instituto de Biología, UNAM (CNIN) y otros 2 en la Colección Entomológica del Instituto de Ecología, A.C. (IEXA). Se examinaron además especímenes adicionales de *L. collaris* procedentes de la Colección Entomológica del Instituto de Ecología, A.C. (IEXA). Dos ejemplares se compararon con el Holotipo y Alotipo de *L. jamesonae*. Para la determinación de los especímenes se utilizaron los trabajos de Delgado (1997) y Soula (2005).

Resultados y Discusión

Lagochile occidentalis sp. nov.

Holotipo macho: longitud del ápice del labro al ápice del pigidio: 19.4 mm; anchura máxima basal y posterior de los élitros 11.00 y 11.2 mm. Coloración dorsal café claro, predominante en élitros y escutelo; cabeza negra con una mancha amarilla posteromedia, sin alcanzar el borde anterior del labro. Pronoto con una mancha amarilla anteromedia, abierta en su parte anterior pero sin alcanzar la base del pronoto, con dos manchas laterales anchas y una línea delgada en la base de color amarillo. Escutelo con una mancha triangular central negra; borde de las mandíbulas, fémures, y tibias, gran parte del metatórax y ventritos abdominales visibles 1-5 y penúltimo tergitoo con tonalidades de color negro; canto ocular negro. Cabeza, pronoto, escutelo, y élitros casi lisos, con puntuación diminuta, esparcida.

Cabeza: clípeo y labro con una escotadura profunda, mucho más marcada en el labro; borde del clípeo con escasas sedas largas, erectas, borde anterior con una franja delgada amarilla; labro con abundantes sedas largas en los bordes interno y externo; ápice del mentón ligeramente sinuado, con abundantes sedas erectas a los lados. Último artejo del palpo maxilar con una leve depresión longitudinal dorsal; anchura interocular (a la altura de la base del canto ocular): 2.8 mm; antena con maza antenal de 2.0 mm, resto de la antena de 2.8 mm; escapo con macrosedas. Pronoto: ángulos anteriores obtusos y redondeados; bordes posterolaterales ligeramente convergentes, borde basal proyectado hacia atrás a la altura del escutelo, ancho de 10.0 mm, largo 4.2 mm; márgenes anteriores y anterolaterales finamente marcados. Escutelo: ancho 4.6 mm y largo de 4.2 mm, con el ápice redondeado. Élitros: moderadamente convexos, levemente aplanados en la región

postescutelar; bordes laterales ligeramente curvos en la parte anterior, con tres grupos de macrosedas a la altura de la base metasternón. Región ventral: con sedas amarillentas, densas y moderadamente largas; primeros cuatro ventritos completamente visibles con una fila transversa de sedas; proceso meso-metasternal ancho, en vista frontal de apariencia triangular, extendiéndose hasta las procoxas y con el ápice redondeado y arqueado dorsalmente en el ápice; sutura meso-metasternal indistinta; metasternón con una línea longitudinal conspicua; ápice del último ventrito denticulado y sinuado mesialmente, con sedas largas; con una línea adicional corta de dentículos pequeños cercana a la parte media. Pigidio: prominente hacia el ápice, ligeramente sinuado pero sin protuberancias conspicuas; superficie con estrías transversas, más profundas y densas hacia los lados, ápice con sedas largas en el tercio apical; último ventrito con una carina preapical transversa, dentículada, escotada en su parte media, con macrosedas en el borde. Patas: protibia tridentada, diente basal más pequeño y más separado que los distales, protarsos 2-4 ligeramente engrosados hacia el ápice, protarsomeros 3-4 escotados, quinto tarsómero con la uña interna bifida, parte interna engrosada y truncada diagonalmente, parte externa aciculada, ligeramente más larga que la parte interna; fémures con 7 espolones lateroapicales; metatrocánter con el ápice agudo y sobresaliendo ligeramente del borde posterior del fémur; metafémures más anchos que la metacoxa. Genital: Parámeros notablemente anchos desde la base, con ligera constricción preapical, con una pequeña proyección meso-apical y un surco mesial en el tercio apical (vista dorsal); ápices casi rectos y agudos (vista lateral) (Figs. 1-6).

Variación: paratipos (16 machos, 10 hembras). Coloración dorsal variable (Figs. 7-12). Algunos especímenes muestran totalidades muy claras y sólo una hembra tiende a ser obscura dorsalmente. Existe variación en la longitud total, distancia interocular, pronoto, escutelo, y anchura de los élitros. Entre machos y hembras: en general las hembras son más grandes (Cuadro 1). Hembras con el último ventrito con dos carinas preapicales transversas, la penúltima dentículada, no escotada, y con macrosedas.

Material tipo: Holotipo ♂: MEXICO: Jalisco: San Sebastián del Oeste, Hacienda Las Tres Carmelitas, bosque tropical subcaducifolio, 20°45'37.4"N, -104°57'59.2"W, 759 msnm, 17.X.19, Oscar D. Hernández L., Guillermo Rodríguez, Carpotrampa, CT-1, 1♂. 29 Paratipos, todos de la misma localidad, excepto: **bosque de encino**, 20°45'19.9"N, -104°58'0.85"W, 837 msnm, 8.IX.19 Oscar D. Hernández L., Guillermo Rodríguez, CT-15, 1♂; **bosque mesófilo de montaña**, 20°45'24.5"N, -104°58'10.1"W 12.VIII.19, 799 msnm, Oscar D. Hernández L., Guillermo Rodríguez, CT-7, 1♂ (CNIN); 20°45'22.8"N, -104°58'11"W, 819 msnm, 16.IX.19, Oscar D. Hernández L., Guillermo Rodríguez, CT-8, 4♂ 1♀; 20°45'22.8"N, -104°58'11"W, 792 msnm, 31.VIII.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 5, 1♀ (IEXA); 20°45'25"N, -104°58'9.1"W, 709 msnm, 31.VIII.19, Oscar D. Hernández L., 20°45'25"N, -104°58'9.1"W, 709 msnm, 3.VIII.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 6, 1♂ (IEXA); 20°45'25"N, -104°58'9.1"W, 709 msnm, Oscar D. Hernández L., Guillermo Rodríguez, CT- 6, 1♂; 20°45'22.8"N, -104°58'11"W, 792 msnm, 17.X.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 10, 1♂; **bosque tropical caducifolio**, 20°45'37.4"N, -104°57'59.2"W, 759 msnm, 6.X.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 1, 1♂, 1♀; 20°45'37.4"N, -104°57'59.2"W, 759 msnm, 16.X.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 1, 5♀(1CNIN, 4CZUG); 20°45'36.5"N, -104°57'59.8"W, 773 msnm, 10.VII.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 2, 2♀; 20°45'36.5"N, -104°57'59.8"W, 773 msnm,

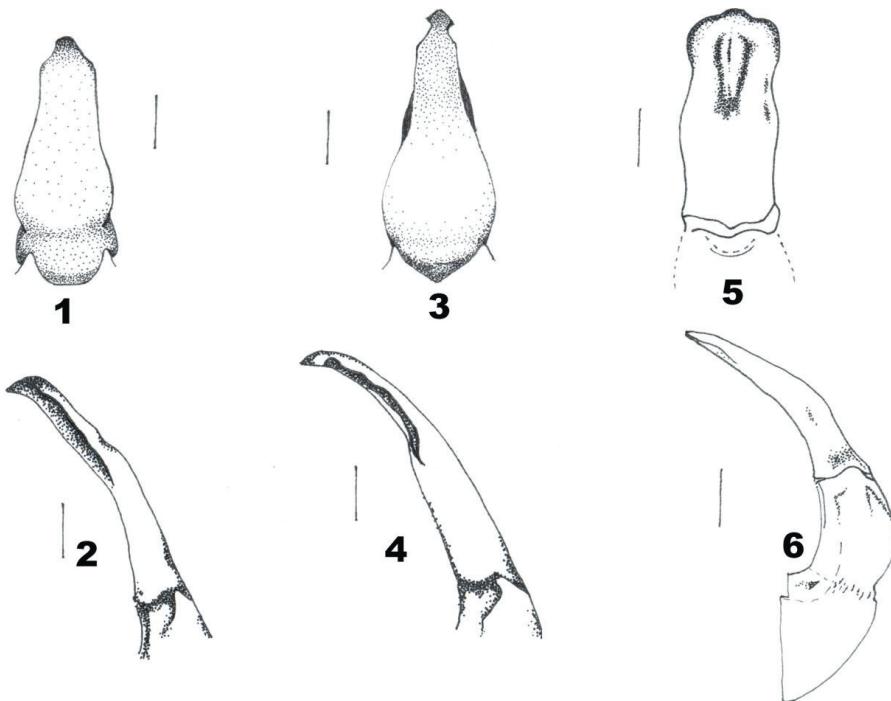


Fig. 1-6. Genital masculino de las especies mexicanas de *Lagochile*. 1, 3, 5. Vista dorsal, 2, 4, 6. Vista lateral. 1-2. *L. jamesonae*. 3-4. *L. collaris*. 5-6. *L. occidentalis*. 1-4. Redibujadas de Delgado (1997).

Fig. 1-6. Male genitalia of Mexican *Lagochile* species. 1, 3, 5. Dorsal view. 2, 4, 6. Lateral view. 1-2 *L. jamesonae*. 3-4. *L. collaris*. 5-6. *L. occidentalis*. 1-4. After Delgado (1997).

26.X.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 2, 1♂; 20°45'36.5"N, -104°57'59.8"W, 773 msnm, 8.IX.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 2, 1♂; 20°45'38.5"N, -104°57'54.8"W, 782 msnm, 10.VIII.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 3, 1♂; 20°45'38.5"N, -104°57'54.8"W, 782 msnm, 17.VIII.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 3, 1♂; 20°45'38.5"N, -104°57'54.8"W, 782 msnm, 3.X.19, Oscar D. Hernández L., Guillermo Rodríguez, CT- 3, 1♂; bosque de pino, 20°45'1.5"N, -104°58'05.8"W, 800 msnm, 8.IX.19, Oscar D. Hernández L., Guillermo Rodríguez, CT-18, 1♂. El holotipo y 25 paratipos se encuentran depositados en la colección CZUG. Dos paratipos, hembra y macho serán depositados en las colecciones del Instituto de Ecología, A.C. y en la Colección Nacional de Insectos (Instituto de Biología, Universidad Nacional Autónoma de México).

Localidad tipo: México: Jalisco: San Sebastián del Oeste, Hacienda Las Tres Carmelitas, bosque tropical subcaducifolio, 20°45'37.4"N, -104°57'59.2"W, 759 msnm.

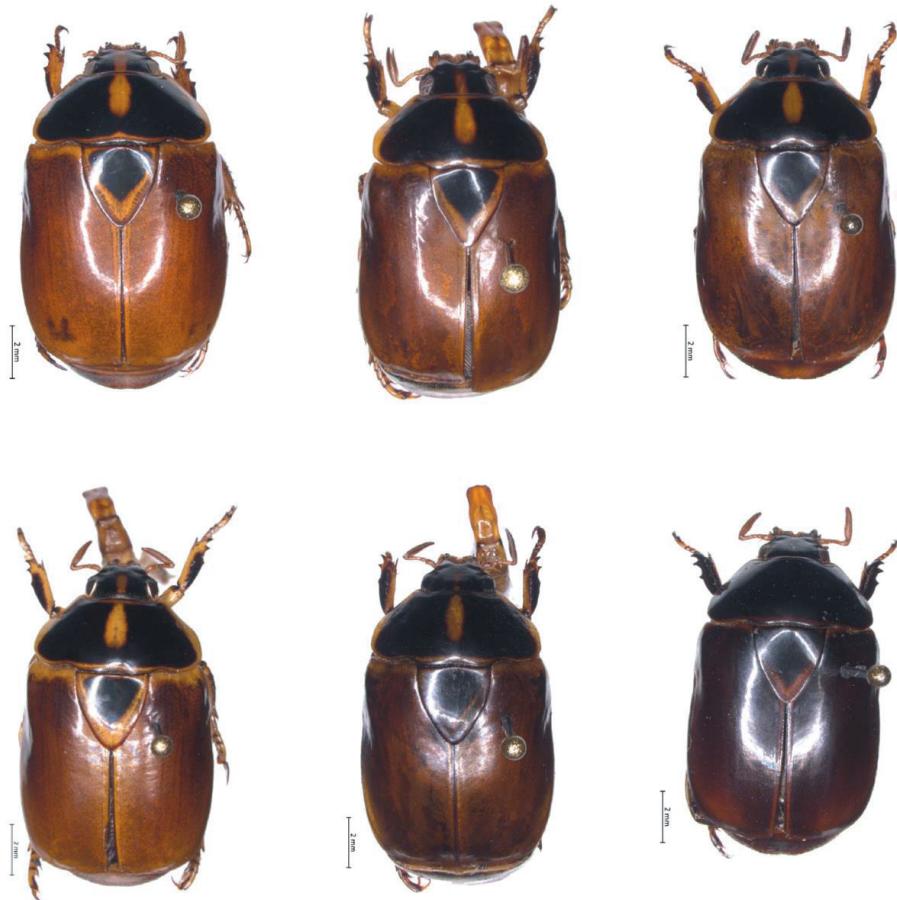


Fig. 7-12. Variación dorsal de la coloración en seis paratipos de *L. occidentalis* sp. nov.

Fig. 7-12. Dorsal color variation in six paratypes of *L. occidentalis* sp. nov.

Etimología: El epíteto específico *occidentalis* hace referencia a la distribución conocida actual para el occidente de México y Jalisco.

Comentarios taxonómicos: Las tres especies de *Lagochile* en México son similares en su apariencia externa, sin embargo, la distinción principal radica en las características del genital (Figs. 1-6) y su distribución hasta ahora conocida: la distribución de las tres especies es alopátrica. Por la vertiente del Pacífico se distribuyen *Lagochile occidentalis* de Jalisco y *L. jamesonae* de Guerrero. *Lagochile occidentalis* se distingue de *L. jamesonae* por la forma de edeago, particularmente en el ápice, además de su distribución geográfica que está restringida al occidente de Jalisco. La coloración de *L. jamesonae* es más pálida en comparación con *L. occidentalis*; en *L. jamesonae* la mancha pronotal central esta completamente

Cuadro 1. Longitud de los Especímenes de la Serie Tipo de *Lagochile occidentalis* sp. nov. de San Sebastián del Oeste, Jalisco, México. Longitud en mm.

Table 1. Length of the Specimens of the Type Serie of *Lagochile occidentalis* sp. nov. from San Sebastián del Oeste, Jalisco, Mexico. Length in mm.

Medidas	Holotipo	♂	♀
Cabeza			
Espacio entre ojos	2.8	2.6-3.0	2.6-2.8
Largo mazo antenal	2.0	1.2-2.1	1.9-2.2
Artejos basales	2.8	1.8	1.6-2.8
Pronoto			
Largo	4.2	3.8-4.4	4.0-4.2
Ancho	10.0	9.0-10.2	9.2-9.8
Escutelo			
Largo	4.2	4.0-4.8	4.0-4.2
Ancho	4.6	3.8-4.4	4.0-4.6
Élitro			
Largo	14.2	13.2-14.0	13.6-15.2
Ancho	11.4	10.5-12.0	11.0-12.0

dividida formando dos manchas o ligeramente fusionada en la base, mientras que en *L. occidentalis* la mancha pronotal está fusionada en la base por una franja ancha.

Distribución: *Lagochile occidentalis* se conoce hasta ahora sólo de la localidad tipo, en el estado de Jalisco.

Ejemplares Adicionales Examinados:

Lagochile collaris Blanchard, 1850

CHIAPAS: G. Halffter, IX.63, G. Halffter leg./ *Chasmodia collaris*, F. Capistran det./ etiqueta amarilla sin datos (1♀); VERACRUZ: Xalapa, Rancho Guadalupe, en suelo, 1430 m, 27 Sept. 1999, M.A. Morón col. (1♀); VERACRUZ: Región de los Tuxtlas, 400-900 m, VII-VIII/06 (2); Puebla, Tlatlauquitepec, Presa la Soledad, 15 Sept. 2013 (2). Este material se encuentra depositado en la Colección particular del Dr. Andrés Ramírez Ponce, Coatepec, Veracruz.

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Referencia Citadas

- Albertoni, F. F., J. Fuhrmann, and S. Ide. 2014. *Lagochile emarginata* (Gyllenhal): Morphology of immature and imago, and biological records (Coleoptera, Scarabaeidae, Rutelinae). Rev. bras. Entomol. 58: 32-46.
<https://doi.org/10.1590/S0085-56262014000100007>

- Blanchard, E. 1850. Catalogue de la Collection Entomologique Classe des Insectes: Ordre des Coléoptères. Première Livraison. Gide et Baudry, Paris.
- Delgado, L. 1997. Una especie nueva de Chasmodia del trópico del Pacífico Mexicano (Coleoptera: Melolonthidae; Rutelinae; Rutelini). Folia Entomol. Mex. 100: 15-21.
- Jameson, M. L., and M. A. Morón. 2001. Descriptions of the larvae of *Chlorota cincticollis* Blanchard and *Chasmodia collaris* (Blanchard) (Sacarabaeidae: Rutelinae: Rutelini) with a key to the larvae of the American genera of Rutelini. Coleopts. Bull. 55: 385-396.
- Morón, M. A. 1997. Capítulo 2. Rutelinae. In M. A. Morón, B. Ratcliffe, and C. Deloya [eds.], Atlas de los Escarabajos de México, Coleoptera: Lamellicornia, Vol. I Familia Melolonthidae. Conabio, SME.
- Reyes-Novelo, E., y M. A. Morón. 2005. Fauna de Coleoptera Melolonthidae y Passalidae de Tzucacab y Conkal, Yucatán, México. Acta Zool. Mex. (n.s) 21: 15-49.
- Soula, M. 2005. Les Coleopteres du Monde 26, 3: Rutelini 2. Revision des Anthicheirina 3. Hillside Books, Canterbury.

***Cionomimus insolens*¹ and *Anthonomus phoradendrae*¹ Associated with Mistletoe, *Phoradendron densus*², at Arteaga, Coahuila, Mexico**

María Paz-Ponce³, Yolanda Rodríguez-Pagaza³, Oswaldo García-Martínez³, Macotulio Soto-Hernández^{4*}, José Ángel Villarreal-Quintanilla⁵, Sergio R. Sánchez-Peña³, and Alberto Flores-Olivas³

Abstract. *Anthonomus phoradendrae* Anderson and *Cionomimus insolens* (Dietz) were collected at Sierra de Zapalinamé, Arteaga, Coahuila, Mexico, from mistletoe plants, *Phoradendron densus* Torr. ex Trel. (Santalaceae), which is hemiparasitic on *Juniperus angosturana* R.P. Adams and *Juniperus flaccida* Schlechtendal at Sierra Madre Oriental.

Resumen. Se recolectaron *Anthonomus phoradendrae* Anderson y *Cionomimus insolens* (Dietz) en la Sierra de Zapalinamé de Arteaga, Coahuila, Mexico, en plantas del muérdago *Phoradendron densus* Torr. ex Trel., de la familia Santalaceae, planta hemiparásita en *Juniperus angosturana* R.P. Adams y *Juniperus flaccida* Schlechtendal en la Sierra Madre Oriental.

Phoradendron densus (Torr.) Eichl (= *P. pauciflorum* Torr.; = *P. saltillense* Trel.) (Santalaceae), commonly known as dense mistletoe, is a hemiparasitic plant on junipers (*Juniperus angosturana* and *Juniperus flaccida*). These are common trees in the Sierra de Zapalinamé, part of the Sierra Madre Oriental at the State of Coahuila, Mexico. *P. densus* frequently causes severe damage to hosts.

A collection was made on 13 September 2019 at 25° 14' 51.8" N, 100° 54' 32" W, 2,025 m above sea level; near Puerto Chapultepec, Arteaga, Coahuila. The site is a forest area with subalpine and submontane scrub vegetation. Several *P. densus* plants were collected from *J. angosturana* trees. Plants were examined on the same date. The mistletoes had whitish, ripe fruit. Ten active Curculionidae adults were observed and collected on plants (Fig. 1). The specimens were sacrificed in ethyl acetate, mounted with entomological pins and identified using keys of Anderson (1994) and Soto et al (2013).

Anthonomus phoradendrae Anderson (nine specimens) and *Cionomimus insolens* (Dietz) (one specimen) (Curculionidae, Curculioninae, Anthonomini) were identified. *Cionomimus insolens* is a new report for the host plant and the State of

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²Santalales: Santalaceae

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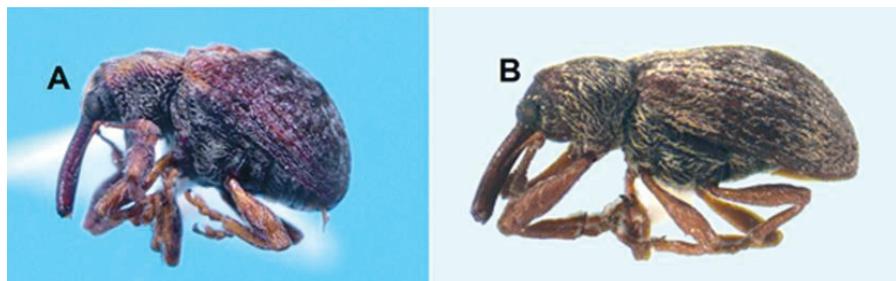


Fig.1. Lateral view of curculionids from *Phoradendron densus* at Sierra de Zapalinamé, Arteaga, Coah. Mex. A) *Cionomimus insolens*, B) *Anthonomus phoradendrae*.

Coahuila, Mexico. All *Cionomimus* species except *C. clarki* are associated with species of *Phoradendron*. *Cionomimus* are distributed from southern Oregon to Baja California, western Mexico, then east through southern Arizona to southwestern Texas, and south through Mexico to Argentina. *A. phoradendrae* was registered by García et. al. (2021), but they did not identify its host plant. We confirmed it is associated with *P. densus*. Most *Anthonomus* have larvae that feed in reproductive structures of the host plant (Burke 1981).

In 1994, Anderson described New World weevil fauna associated with the parasitic plant family Viscaceae (now in Santalaceae), this fauna consisting of 24 species in eight genera of four tribes of Curculionidae, including three new species of *Anthonomus* from southern Mexico: *A. brachyrhinus*, *A. guerreroensis*, and *A. phoradendrae*; also, five new species of *Cionomimus*: one for Venezuela, *C. clarki* (not associated with *Phoradendron*) and the others from Mexico: *C. burkei*, *C. grossus*, *C. obrieni*, and *C. woodi*. Anderson (1997) described five new species of Anthonomini associated with *Phoradendron*: three new species of *Smicraulax* Pierce (*S. nigrinus*, *S. otidocephaloides* and *S. ephippiatus*) and two new species of *Cionomimus*: *C. hansonii* from Costa Rica (not associated with *Phoradendron*) and *C. bimaculatus* from Panama. There are 11 species in the genus.

In Mexico, the host plant *P. densus* is distributed in the States of Baja California, Chihuahua, Coahuila, Nuevo León, San Luis Potosí, and Sonora. The genus *Juniperus* is distributed in most montane areas of the country (Villaseñor 2016). These weevil species could be present in these states also.

References Cited

- Anderson, R. S. 1994. A review of New World weevils associated with Viscaceae (mistletoes [in part] including descriptions of new genera and new species (Coleoptera: Curculionidae). J. Nat. Hist. 28: 435-492.
- Anderson, R. S. 1997. New species and new records of *Smicraulax* Pierce 1908 and *Cionomimus* Marshall 1939 from Central America (Curculionidae Curculioninae Anthonomini), Tropical Zoology 10: 255-270. DOI: 10.1080/03946975.1997.10539341

- Burke, H. R. 1981. Review of the genus *Cionomimus* Marshall with descriptions of two new species (Coleoptera: Curculionidae). Southwest. Entomol. 6: 174-183.
- García-Martínez, O., M. Soto-Hernández, S. Ordaz-Silva, and D. S. Ordaz-Ontiveros. 2021. Weevils (Coleoptera: Curculionidae) of Coahuila de Zaragoza, México. Southwest. Entomol. 46: 947-956.
- Soto, H. M., R. W. Jones, and C. P. Reyes. 2013. A key to the Mexican and Central America Genera of Anthonomini (Curculionidae, Curculioninae), Zookeys 260: 31-47.
- Villaseñor, R. J. L. 2016. Check list of the native vascular plants of México. Rev. Mex. Biodiversidad 87: 559-902.

Poblaciones del Picudo del Chile *Anthonomus eugenii*¹ en Puebla, México**Populations of the Pepper Weevil *Anthonomus eugenii*¹ in Puebla, Mexico**

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Resumen. Se reporta la fluctuación de picudo del chile, *Anthonomus eugenii* Cano, en tres municipios de Puebla, México, la cual se observó desde el inicio de la floración en abril hasta post cosecha en diciembre, con la mayor población en agosto. El monitoreo directo e indirecto a través de trampas cebadas con feromonas fueron igualmente útiles para detectar las primeras poblaciones de *A. eugenii*.

Abstract. Fluctuation in abundance of pepper weevil, *Anthonomus eugenii* Cano, was reported from three municipalities of Puebla, Mexico, observed from the beginning of flowering in April until post-harvest in December with most abundance in August. Direct and indirect monitoring with pheromone-baited traps were equally useful in early detection of pepper weevil.

La producción de chile (*Capsicum annuum* L.) en México es importante y lo hace el segundo país productor de >3 millones de toneladas anuales (SIAP 2021). *Myzus persicae* Sulzer, *Aphis gossypi* Glover, *Frankliniella occidentalis* Pergande, *Trips tabaci* Lindeman, *Liriomyza trifolii* Burgess, *Spodoptera exigua* Hübner, y picudo del chile, *Anthonomus eugenii* Cano, son sus plagas principales (Jiménez 2014). *Anthonomus eugenii* la plaga más importante que puede causar pérdidas del 100% si no se realizan acciones de control a tiempo (Avedaño et al. 2015). Los mismos autores señalan que la medida de control más utilizada por los productores de Sinaloa es la aplicación de los insecticidas clorpirifos, malatión, oxamil, zeta cipermetrina, y thiametoxam. Este último insecticida ha registrado la mayor tolerancia en La Cruz de Elota, Sinaloa.

El censo se realizó en los municipios de Tepexi de Rodríguez, Ixcaquixtla, y Coyotepec, Puebla, México entre el 25 de febrero al 27 de julio, y del 11 de agosto al 07 de diciembre del 2021. Se seleccionaron seis parcelas y en cada una se realizaron muestreos directos, que consistieron en la recolecta manual de adultos, flores y frutos que presentaban síntomas de la presencia del picudo. En cada parcela se seleccionaron 15 puntos al azar, y en cada punto se delimitó 1 m² donde se ubicaron seis plantas. En éstas se observó follaje, flores, y frutos para determinar la

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presencia de *A. eugenii* (Anovel et al. 2015). El periodo de los muestreos indirectos se realizó del 08 de abril al 11 de agosto de 2021, y del 26 de agosto al 23 de enero del 2022. Se utilizaron 39 ‘kits’ de trampas comerciales Pherocon Pew® (TRÉCÉ, IA), las cuales incluyen una trampa amarilla con adherente a base de poliisobutileno (15 cm de ancho x 31 cm de largo), un atrayente alimenticio o kairomona ((E) -3,7-dimethyl-2,6-octadienoic acid (geranic acid)), y una feromona de agregación Pherocon que contiene (Z)-2-(3,3-dimethylcyclohexylidene) ethanol, (E)-2-(3,3-dimethylcyclohexylidene) ethanol, (Z)-(3,3-dimethylcyclohexylidene) acetaldehyde, (E)-(3,3-dimethylcyclohexylidene) acetaldehyde y (E)-3,7-dimethyl-2,6-octadien-1-ol (geraniol). Se colocó una trampa en el centro de cada parcela utilizada en el muestreo directo, sobre una estaca de madera a 10 y 60 cm del suelo (dependió de la altura de la planta), posteriormente se colocaron los dos atrayentes en la parte superior de la trampa amarilla. Las trampas amarillas se recogieron cada 15 días y se contó el número de picudos. Los datos de relación entre cada tipo de monitoreo se compararon mediante el test χ^2 de Pearson ($P > 0.05$).

En la zona de estudio realizan aplicaciones preventivas de plaguicidas sintéticos para el manejo de plagas y enfermedades, siendo esto un factor que afecta la fluctuación poblacional de la plaga. Sin embargo, al no aplicar los daños y las pérdidas serían cuantiosas. Aun así los primeros picudos se observaron de 70 a 80 días después de la plantación a finales de abril en etapa de floración y fructificación. El mayor número de huevos, larvas, pupas y adultos se encontró en julio con 889 picudos, y 381 larvas (Fig. 1). En septiembre se detectaron en la etapa de floración (398 adultos). La población aumentó a 730 en octubre, disminuyendo en noviembre y diciembre a 343 adultos y 33 larvas.

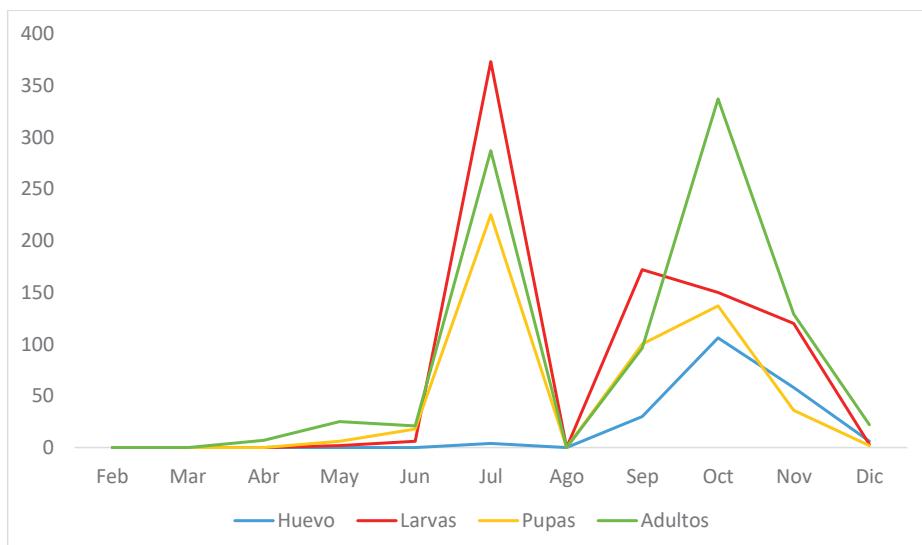


Fig. 1. Número de huevos, larvas, pupas, y adultos de *Anthonomus eugenii* en chile en Puebla, México.

Fig. 1. Number of eggs, larvae, pupae, and adults of *Anthonomus eugenii* in pepper in Puebla, México.

Se observaron los primeros adultos en la segunda quincena de abril, y larvas y pupas para la primera quincena de mayo y en julio los primeros huevos, además que fue en el mismo mes donde se encontró la mayor población: 4 huevos, 373 larvas, 225 pupas, y 287 adultos. Para el segundo ciclo agrícola, en septiembre se observaron los primeros 30 huevos, 172 larvas, 100 pupas, y 96 adultos, y la mayor población fue en octubre con 106 huevos, 150 larvas, 137 pupas, y 337 adultos.

En base a los resultados la fluctuación poblacional del picudo del chile presentó dos picos de población, el primero en julio en donde predomina la presencia de larvas, el declive que se observa en el mes de agosto se debe a que no se realizó ningún muestreo directo debido a que los predios solo presentaban residuos de cosecha. Sin embargo, para el septiembre se observó el crecimiento de la plaga. El segundo pico poblacional se presentó en octubre con mayor incidencia de adultos.

A diferencia del muestreo directo (Fig. 2), en estos resultados se observa un pico de población en agosto. Esto se puede deber a que en la fecha de recolección de la trampa los predios presentaban residuos de cosecha y no se estaba realizando ningún tipo de manejo. En consecuencia, las poblaciones del picudo del chile aumentaron debido a que los productores no realizan un manejo post cosecha, permitiendo que la plaga cuente con hospederos para su reproducción. Ante esto se recomienda realizar manejo de post cosecha para evitar el aumento de las poblaciones del picudo del chile.

Comparando las dos técnicas de muestreo la mayor población de picudos del chile se observó en agosto mediante muestreo indirecto, lo que puede ser la causa por la que en el segundo ciclo agrícola se detectó un mayor número de picudos al inicio del ciclo de la plantación. En diciembre se finalizaron las cosechas del segundo ciclo agrícola, pero las poblaciones de picudo se siguieron manteniendo hasta enero aun sin cultivos en pie. Estos resultados demuestran la utilidad de las trampas con

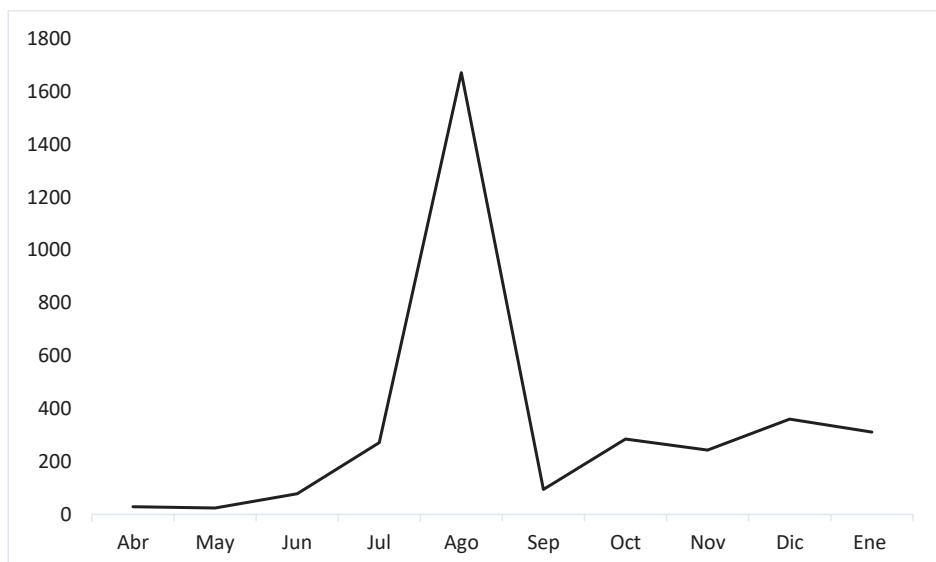


Fig. 2. Número de adultos de *Anthonomus eugenii* en chile en Puebla, México.
Fig. 2. Number of adults of *Anthonomus eugenii* in chili in Puebla, México.

feromonas de agregación y atrayente alimenticio tanto para el monitoreo de la plaga como para el manejo. Además, se determinó que los picudos se encuentran activos aun después de la cosecha como se puede observar en la Fig. 2, siendo un foco de infestación entre ciclo y ciclo. Los resultados de este trabajo concuerdan con lo descrito por Sparks et al. (2022), quienes mencionan que las posibilidades de reinfección de nuevas siembras de chile se deben a: 1) el uso de plántulas infestadas y 2) el traslape de las plantaciones. No se observó hibernación de *A. eugenii* en Puebla, ya que la población estuvo activa en diciembre y enero, lo que hace probable que el traslape de generaciones se deba a que en Puebla se tienen plantas de chile todo el año.

La efectividad del muestreo directo e indirecto no muestran diferencias ($P = 0.3336$), por lo que los dos métodos son una herramienta confiable para la detección inicial de las poblaciones del picudo. Esta fluctuación poblacional está directamente relacionada con la etapa fenológica del cultivo, así como por la calidad y cantidad del alimento disponible, debido a que la plaga se observa al inicio de la floración y se mantiene durante la floración y fructificación.

Agradecimiento

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Referencias Citadas

- Anovel, B., A. C. Vidal, G. M. Román, y H. Masachika. 2015. Dinámica poblacional del picudo del Ají *Anthonomus eugenii* (Coleoptera: Curculionidae). Ciencia Agropecuaria 22: 1-15.
- Avendaño, M. F., T. S. Parra, M. J. L. Corrales, y P. P. Sánchez. 2015. Resistencia a insecticidas en tres poblaciones del picudo del chile (*Anthonomus eugenii* Cano) en el estado de Sinaloa, México. Fitosanidad 19: 193-199.
- Jiménez, B. J. 2014. Manual técnico de: Plagas y Enfermedades en Solanáceas. Gowan Mexicana SAPI de C.V. 395.
- SIAP (Servicio de Información Agroalimentaria y Pesquera). 2021. Panorama Agroalimentario 2021. https://nube.siap.gob.mx/gobmx_publicaciones_siap/pag/2021/Panorama-Agroalimentario-2021 Recuperado el 2 Febrero 2022.
- Sparks, A. N., T. R. Weredyk, T. Torrance, J. Shealey, S. Hollifield, W. Gay, J. Kichler, and J. Hand. 2022. Overwintering of *Anthonomus eugenii* (Coleoptera: Curculionidae) in southern Georgia. J. Entomol. Sci. 57: 123-128.

Interacciones entre Hormigas¹ y la Mosca Negra, *Aleurocanthus woglumi*², sobre Cultivos de Palmilla *Chamaedorea radicalis*³**Interactions between Ants¹ and Blackfly, *Aleurocanthus woglumi*², in Palmilla Crops *Chamaedorea radicalis*³**

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Resumen. Se presenta el primer registro de interacciones entre dos especies de hormigas nativas y dos especies introducidas en México con la mosca negra (*Aleurocanthus woglumi* (Ashby, 1915)), especie introducida de importancia económica en cítricos en México, en un cultivo *in-situ* de palmita (*Chamaedorea radicalis* Mart.) en Tamaulipas, México. Se reportan cinco observaciones de comportamientos de cuidado hacia las moscas.

Abstract. The interactions between two native and two introduced ant species with the introduced black citrus pest (*Aleurocanthus woglumi* (Ashby, 1915)), was recorded over palmilla (*Chamaedorea radicalis* Mart.) plants at Tamaulipas, Mexico. Five observations of ant care behavior toward the fly pest are reported.

Las hormigas establecen relaciones estrechas y frecuentes con gran variedad de especies de plantas, hongos y particularmente con otros insectos fitófagos (Stadler y Dixon 2008). Una de estas interacciones es con especies del orden Hemiptera (en especial del suborden Sternorrhyncha). Éstos succionan la savia de las plantas y secretan azúcares (melaza) que son consumidas por las hormigas a cambio de brindar protección y cuidados a los hemípteros (Delabie y Fernández 2003, Renault et al. 2005, Stadler y Dixon 2005, Ross y Shucker 2009, Ivens 2015). Esta asociación entre hormigas y hemípteros se conoce como trofobiosis, y puede causar repercusiones en los cultivos permitiendo un crecimiento exponencial de las plagas (Ferrer et al. 1999, Styrsky y Eubanks 2007, Miller y Ben-Dov 2013). En México se han realizado escasos estudios sobre la asociación hormiga-hemíptero en especies vegetales de importancia económica (Cuautle et al. 1999, Perfecto y Vandermeer 2006). *Chamaedorea radicalis* es una especie endémica de palmera del sotobosque en Nuevo León, San Luis Potosí y Tamaulipas México, cosechada habitualmente por la población local para vender las hojas pinnadas a los mercados

¹Hymenoptera: Formicidae

²Hemiptera: Aleyrodidae

³Arecales: Arecaceae

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internacionales (Jones y Gorchov 2000). Después del 2000 se han utilizado estrategias de manejo de poblaciones silvestres y el establecimiento de plantaciones comerciales para reducir la explotación sobre las poblaciones naturales (Granados-Sánchez 2004, Montelongo-Ruiz et al 2020), lo que ha llevado a un aumento de su producción. Adicionalmente en estas zonas se cultivan huertos frutales que incluyen cítricos.

Con el objetivo de observar la interacción hormiga-hemíptero, en el municipio de Gómez Farías, Tamaulipas, se establecieron 3 sitios de cultivos agroforestales de palmilla (*Chamaedorea radicalis* Mart.) separados por al menos un kilómetro de distancia y en elevaciones de 150-350 m s. n. m. Cada sitio midió 800 m². Los cultivos de palmilla presentan diferentes especies nativas, debido a que para su crecimiento es necesaria la sombra de acahuales y bosques (Quero 1994), entre ellas: *Bursera simaruba* (L.) Sarg. 1890, *Cedrela odorata* L. 1753, y *Lysiloma divaricatum* (Jacq.) J.F.Macbr. El Sitio 1 (23°04'42.5" N, 99°09'40.9" W) está rodeado por selva baja subcaducifolia; Sitio 2 (23°01'6.2" N, 99°09'23.4" W) se encuentra flanqueado por zonas de pastoreo, cultivos de naranja (*Citrus sinensis* [L.] Osbeck, variedad Valenciana) y caña (*Saccharum officinarum* L.); Sitio 3 (23°04'29.4" N, 99°08'39" W) se ubica junto a viviendas a 10 metros de distancia de la carretera. La densidad relativa vegetal de la palmilla en los tres sitios fue 80%. El 96% de los propietarios indican que implementan un manejo cultural (arranque, quema o corte frecuente), mientras que 4% utilizan fertilizantes y/o pesticidas en sus cultivos. En cada sitio se establecieron 5 transectos de 25 m y cada 5 m se designó una subestación de muestreo (Lozano et al. 2013). Los muestreos se realizaron en abril, agosto, y noviembre del 2019. Los datos de observación de las especies de hormigas y la mosca negra se documentaron por medio de videos (formato .MP4, 1280x720, 29 f/s) a partir de la revisión de diez plantas al azar por subestación examinando el envés de las frondas de las palmillas (2,250 palmillas) de 09:00-17:00 h. Por medio de colecta directa se obtuvieron los ejemplares de formícidos y *A. woglumi* para su preservación en alcohol al 70%. La identificación de hemípteros se realizó con las claves en McKenzie (1960), Howell y Williams (1976), Martín (1987), Myartseva et al. (2016), y Ramos y Caballero (2017). Para la identificación de género de las hormigas se utilizaron las claves de Mackay y Mackay (1989) y para la identificación a especie se utilizaron claves especializadas por Cuezzo et. al. (2015), Longino (2003), y Ortiz et. al. (2019).

Se registraron cinco interacciones entre hormigas obreras (102 individuos) y moscas negras ((*Aleurocanthus woglumi* (Ashby, 1915)) (202 individuos). En todos los casos en donde se observó la presencia de moscas negras, se registraron mediante grabaciones, comportamientos de cuidado (Fig. 1). Estas observaciones únicamente se presentaron en el Sitio 2 en dónde se utilizó exclusivamente un manejo cultural. Aunque no se observó que los formícidos defendieran a los individuos de *A. woglumi* de sus depredadores, si se registró un aseo y melado por lo que ambos grupos potencialmente se pueden beneficiar uno del otro. Se reporta la interacción con las siguientes especies de hormigas: *Brachymyrmex patagonicus* Mayr, 1868 (2 obreras), *Crematogaster curvispinosa* Mayr, 1862 (41 obreras), *Monomorium floridola* Jerdon, 1851 (12 obreras), y *Wasmannia auropunctata* (Roger, 1863) (47 obreras), estas dos últimas consideradas como especies introducidas para México.

Las hormigas reportadas son dominantes asociadas a recursos con alto contenido de carbohidratos como las secreciones producidas por hemípteros (Brandão et al. 2012). Se sabe que *A. woglumi* fue introducida a México mediante

el comercio de frutas (Arredondo et al. 2015) y se ha reportado en Tamaulipas como una plaga importante en cítricos (Rossini et al. 2015). El único sitio (2) para este estudio en donde se observaron estas interacciones se sitúa junto a cultivos de naranja, por lo que las asociaciones de las hormigas - mosca negra y el cultivo de palmillas podrían favorecer la protección del hemíptero, y su posterior dispersión a las áreas citrícolas. Se sugiere realizar estudios en cuanto a la función de *Chamaedorea radicalis* como reservorio de *A. woglumi* y el impacto en el crecimiento poblacional de esta especie en ausencia y presencia de hormigas.

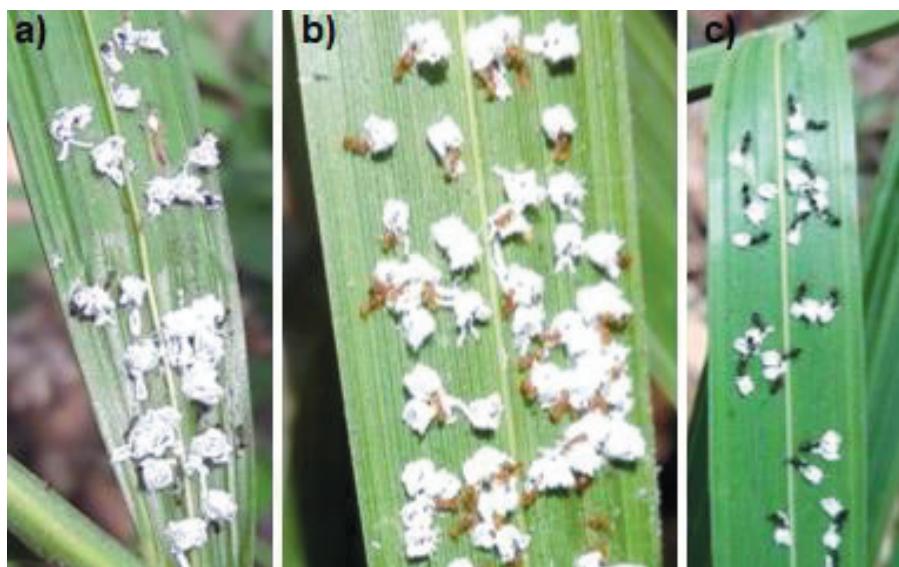


Fig. 1. Interacción entre hormigas y *Aleurocanthus woglumi* en el envés de las hojas de la palmilla *Chamaedorea radicalis*. a) *Monomorium floricola*, b) *Wasmannia auropunctata*, c) *Crematogaster curvispinosa*.

Fig. 1. Associations between ants and *Aleurocanthus woglumi* on the underside of palmilla leaves *Chamaedorea radicalis*. a) *Monomorium floricola*, b) *Wasmannia auropunctata*, c) *Crematogaster curvispinosa*.

Referencias Citadas

- Arredondo-Bernal, H. C., M. A. Mellín-Rosas, y E. Jiménez-Jiménez. 2015. Mosca Prieta de los Cítricos, *Aleurocanthus woglumi* (Hemiptera: Aleyrodidae), pp. 333-346. En H. C. Arredondo-Bernal y L. A. Rodríguez del Bosque [eds.], Casos de Control Biológico en México. Vol. 2. Printing arts Mexico S. de R. L. de C. V. México, DF.
- Brandão, C. R., R. R. Silva, and J. H. Delabie. 2012. Neotropical ants (Hymenoptera) functional groups: nutritional and applied implications, pp. 213-236. In Insect Bioecology and Nutrition for Integrated Pest Management,

- Cuautle, M., V. Rico-Gray, J. G. García-Franco, J. López-Portillo, and L. B. Thien. 1999. Description and seasonality of a Homoptera-ant-plant interaction in the semiarid Zapotitlán Valley, Puebla, Mexico. *Acta Zool. Mex.* (n.s.) 78: 73-82.
- Cuezzo, F., L. A. Calcaterra, L. Chifflet, and P. Follett. 2015. *Wasmannia* Forel (Hymenoptera: Formicidae: Myrmicinae) in Argentina: systematics and distribution. *Sociobiol.* 62: 246-265.
- Delabie, J. H. C., y F. Fernández. 2003. Relaciones entre hormigas y "Homópteros" (Hemiptera: Sternorrhyncha y Auchenorrhyncha), pp. 181-197. En F. Fernández [ed.], *Introducción a las Hormigas de la Región Neotropical*. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt. Bogotá DC., Colombia. ISBN: 958-8151-23-6.
- Ferrer, M. T. M., R. Palacios, y X. Cerdá. 1999. Composición, abundancia y fenología de las hormigas (Hymenoptera: Formicidae) en campos cítricos de Tarragona. *Bol. Sanid. Veg. Plagas* 25: 229-240. Sitio red: <https://dialnet.unirioja.es/servlet/articulo?codigo=114179>
- Granados-Sánchez, D., M. A. Hernández-García, G. F. López-Ríos, y M. Santiago-López. 2004. El cultivo de palma camedor (*Chamaedorea* sp.) en sistemas agroforestales de Cuichapa Veracruz. *Rev. Fitotec.* 27: 233-241.
- Howell, J. O., and M. L. Williams. 1976. An annotated key to the families of scale insects (Homoptera: Coccoidea) of America, north of Mexico, based on characteristics of the adult female. *Ann. Entomol. Soc. Am.* 69: 181-189.
- Ivens, A. B. 2015. Cooperation and conflict in ant (Hymenoptera: Formicidae) farming mutualisms: a review. *Myrmecol. News* 21: 19-36.
- Jones, F. A., and D. L. Gorchov. 2000. Patterns of abundance and human use of the vulnerable understory palm, *Chamaedorea radicalis* (Arecaceae), in a montane cloud forest, Tamaulipas, Mexico. *Southwest. Nat.* 45: 421-430.
- Longino, J. T. 2003. The *Crematogaster* (Hymenoptera, Formicidae, Myrmicinae) of Costa Rica. *Zootaxa* 151: 1-150.
- Lozano, J. C. A., Á. M. A Cardona, y P. C. de Ulloa. 2013. Hormigas en cultivos de naranja (Citrus sinensis L. Osbeck) de la costa Caribe de Colombia. *Biota Colomb.* 14: 13-19.
- Mackay, W. P., y E. Mackay. 1989. Clave de los géneros de hormigas en México (Hymenoptera: Formicidae), pp. 1-82. En *Memorias del II Simposio Nacional de Insectos Sociales*. SME-CIEAMAC, Oaxtepec, Morelos, México.
- Martin, J. H. 1987. An identification guide to common whitefly pest species of the world (Homopt. Aleyrodidae). *Trop. Pest Manag.* 33: 298-322.
- McKenzie, H. 1960. Taxonomic study of California mealybugs, with descriptions of new species (Homoptera: Coccoidea: Pseudococcidae). *Hilgardia* 29: 681-770.
- Miller, D. R., and Y. Ben-Dov. 2013. EscaleNet: database of the scale insects of the world. <http://www.sel.barc.usda.gov/scalenet/scalenet.htm>.
- Montelongo-Ruiz, G., Y. D. R. Moreno-Ramírez, M. C. Juárez-Aragón, N. Niño-García, R. I. Torres-Acosta, and J. A. Torres-Castillo. 2020. Tentative identification of phytochemicals and antioxidant activities during fruit-ripening on *Chamaedorea radicalis* Mart. *Phyton*. 89: 361-373.
- Myartseva, S. N., E. Ruiz-Cancino, y J. M. Coronado-Blanco. 2016. Conocimiento actual de los enemigos naturales (Hymenoptera: Aphelinidae) de escamas armadas (Hemiptera: Diaspididae) en México y descripción de una especie nueva de *Coccobius*. *Acta Zool. Mex.* 32: 81-89.

- Ortiz Sepulveda, C. M., B. Van Bocxlaer, A. D. Meneses, and F. Fernández. 2019. Molecular and morphological recognition of species boundaries in the neglected ant genus *Brachymyrmex* (Hymenoptera: Formicidae): toward a taxonomic revision. *Org. Divers. Evol.* 19: 447-542.
- Perfecto, I., and J. Vandermeer. 2006. The effect of an ant-hemipteran mutualism on the coffee berry borer (*Hypothenemus hampei*) in southern Mexico. *Agric. Ecosyst. Environ.* 117: 218-221.
- Quero, H. J. 1994. Flora de Veracruz. Fascículo No 81. *Palmae*. Instituto de Ecología, A.C., Xalapa, Veracruz, México.
- Ramos, A. A., and A. Caballero. 2017. Diaspididae on *Citrus* spp. (Rutaceae) from Colombia: new records and a taxonomic key to their identification. *Rev. Fac. Nac. Agron. Medellín* 70: 8139-8154.
- Renault, C. K., L. M. Buffa, and M. A. Delfino. 2005. An aphid-ant interaction: effects on different trophic levels. *Ecol. Res.* 20: 71-74.
- Ross, L., and D. M. Shuker. 2009. Scale insects. *Curr. Biol.* 19: R184-R186.
- Rossini, M. N., J. P. Agostini, y D. M. Dummler. 2015. Plagas cuarentenarias de frutales de la República Argentina. Avances en los resultados. Instituto Nacional de Tecnología Agropecuaria. http://inta.gob.ar/sites/default/files/inta_plagas-cuarentenarias-de-frutales-de-la-republica-argentina.pdf
- Sánchez-Ramos, G., P. Reyes-Castillo, A. Mora Olivo, y J. G. Martínez-Ávalos. 2010. Estudio de la herbivoría de la palma camedor (*Chamaedorea radicalis*) Mart., en la Sierra Madre Oriental de Tamaulipas, México. *Acta Zool. Mex.* 26: 153-172.
- Stadler, B., and A. F. G. Dixon. 2005. Ecology and evolution of aphid-ant interactions. *Annu. Rev. Ecol. Evol.* 36: 345-372.
- Stadler, B., and T. Dixon. 2008. Mutualism: ants and their insect partners. Cambridge University Press, Cambridge.
- Styrsky, J. D., and M. D. Eubanks. 2007. Ecological consequences of interactions between ants and honeydew-producing insects. *Proc. Royal Soc. B* 274: 151-164.

Instructions for Preparation of Manuscripts

Manuscripts submitted for consideration for publication in the *Southwestern Entomologist* should report results of entomological research in the southwestern United States or Mexico or should report results of studies on entomological topics relevant to the region, which may be done elsewhere, provided such results are geographically applicable. Manuscripts that report results of routine laboratory or field experiments for which the primary purpose is gathering baseline data or those that report results of a continuous evaluation program such as preliminary pesticide evaluation, species lists with no supporting biological data, or preliminary plant resistance evaluations are not acceptable. However, reports of experiments with insecticides, acaricides, and microbials are acceptable if they are comprehensive and include data related to economics, resistance, toxicology, management, or other broad subject areas. Bibliographies will not be published in *Southwestern Entomologist*.

Scientific Notes can pertain to observations on new hosts, geographic records, and new techniques. Manuscript preparation and review are the same as those for a regular article. Scientific Notes should not exceed four, single-spaced pages and may contain a single figure or table. The References Cited section is the only section that should be identified by a No. 1 heading.

A section entitled "Perspectives" is available for contributions of general interest with an emphasis on papers of a theoretical or historical nature. Because of the nature of Perspectives articles, more tolerance for speculation is acceptable than in regular research articles.

Supplements, which consist of a collection of papers on a similar topic or large monographic articles, may be published periodically. Guidelines for preparation of Supplements are the same as those for regular journal articles. Anyone wishing to develop a supplement should contact the Editor.

It is imperative that the manuscript be prepared in proper journal style. The Editor reserves the right to return or reject a manuscript that will not reproduce clearly or is unsatisfactory for other reasons. Publication charges per printed page or any portion thereof will be \$20 for members and \$25 for non-members. Price quotations on pages printed in color will be made upon request.

The following instructions must be adhered to:

1. Manuscripts should be submitted electronically as Word® files to the Editors [SWEntomologist@gmail.com (English language) or Carlos.Blanco1206@gmail.com (Spanish language)].
2. Letter-sized pages of 8.5 x 11 inches should be used. Except for the margin at the top of the first page, the margins at the top and bottom of each page should be 0.65 inch, with 1.15 inch on the left and right sides of each page.
3. The font should be Arial 12-point. The title should be left justified and in bold font with only the first letter of the main words in upper-case letters on no more than two lines. Leave two line spaces between the title and the author's name(s). The author's name(s) should be left-justified on the page with only the first letters of the name(s) capitalized. Leave one line space between the author's name(s) and the address which also is left justified. Use footnotes in Arial 10-point at the bottom of the page if more than one address for authors is required. The authors should provide e-mail addresses.
4. If insect names (common or scientific) are mentioned in the title, order and family names should be listed as a footnote in Arial 10-point at the bottom of the first page.

Scientific names throughout the manuscript should be in italics and include the author of the species the first time they are used in both the abstract and the text.

5. The text in Arial 12-point should be double-spaced. Both left and right margins should be justified. Do not hyphenate a word at the end of a line. Except for the Abstract, each paragraph should be indented by a half-inch tab space, but not separated by a line space.
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11. Acknowledgments should be written as text and placed before the References Cited section. The word Acknowledgment is singular and centered in bold font with the first letter in upper case.
12. References should be indicated in the text by author and year not separated by a comma. The words References Cited in bold font with the first letter of each word in upper-case should be centered above the reference list at the end of the manuscript. All citations should contain the initials and names of all authors. The complete title of the reference should be given. Titles of journal articles and book chapters are to have only the first letter of the first word capitalized. The title of a book chapter should be followed by the page numbers. The editors of a book should precede the book title; the first letters of major words of a book title should be capitalized. Page numbers of a journal article are one space after the colon following the number of the journal. Do not include issue numbers. Do not use *Ibid* or blank spaces for repetition. The Internet link for the reference may be listed when available.
13. Abbreviations should follow *Scientific Style and Format: The CSE Manual for Authors, Editors, and Publishers*.

Instrucciones para la Preparación de Manuscritos en Español

Manuscritos sometidos a consideración deben reportar resultados de investigaciones entomológicas llevadas a cabo en el suroeste de los Estados Unidos o en México, o reportar resultados de estudios relevantes a esta región, llevados a cabo en otras partes, siempre y cuando estos resultados sean aplicables en dicha zona geográfica. No son aceptables manuscritos que reporten resultados de trabajos rutinarios de laboratorio o de campo, cuyo propósito principal haya sido recolectar información básica, o que reporten resultados de un programa de evaluación continua tales como la evaluación preliminar de plaguicidas, listas faunísticas carentes de datos biológicos, o pruebas preliminares de resistencia vegetal. Sin embargo, reportes de experimentos con insecticidas, acaricidas e insecticidas microbiales son aceptables para su publicación, siempre y cuando sean comprensivos y estén relacionados con aspectos económicos, de resistencia, toxicológicos, manejo, u otros temas de interés amplio. *Southwestern Entomologist* no publicará bibliografías.

Las Notas Científicas pueden describir observaciones de nuevos hospederos, registros geográficos, y nuevas técnicas. Las instrucciones para la preparación de manuscritos y el procedimiento de revisión son iguales a las de artículos regulares. Las Notas Científicas no deberán exceder cuatro páginas a espacio sencillo y pueden contener una sola figura o cuadro. La única sección que debe ser identificada con el encabezado primario (No. 1) es la de Referencias Citadas.

Una sección titulada "Perspectivas" está disponible para contribuciones de interés general con énfasis en trabajos teóricos o de naturaleza histórica. Debido a la naturaleza de estos artículos, se tiene una mayor tolerancia a la especulación que en artículos regulares de investigación.

Los Suplementos, que consisten en colecciones de artículos con un tema común o monografías extensas, podrán ser publicados periódicamente. Las instrucciones para la preparación de estos Suplementos son iguales a las de los artículos regulares. Cualquier persona interesada en desarrollar un Suplemento deberá comunicarse con la/el Editor(a).

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2. Deben usarse páginas de 8.5 pulgadas (21.6 cm) de ancho por 11 pulgadas (27.9 cm) de largo. Mantener márgenes de 0.65 pulgadas (1.6 cm) arriba y abajo de cada página, y de 1.15 pulgadas (2.9 cm) a cada lado.
3. Use el tipo de letra Arial de 12 puntos. El título debe ser alineado a la izquierda, en negrillas (**bold**), usando mayúsculas solo en la primera letra de cada palabra principal, y no debe ocupar más de dos renglones. Se deben dejar dos espacios entre el título y el nombre del autor o autores. El nombre del autor o autores debe alinearse a la izquierda y escribirse usando mayúsculas solo en la primera letra del nombre y apellido(s). Se debe dejar un espacio entre el nombre del autor y la dirección postal y electrónica, que también deben ser alineadas a la izquierda. Use

- notas al pie de página en Arial de 10 puntos si es que hay más de una dirección de los autores; los que deben proveer su dirección electrónica.
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 8. Los pies de página, los cuadros y sus leyendas, y las leyendas de las figuras deben escribirse a espacio sencillo. Arial de 10 puntos debe usarse para palabras escritas en sobrescrito y subíndice.
 9. Los cuadros deben incluirse como parte del texto, pero de tal forma que se evite confusión con el resto del texto; es decir, dejando al menos dos espacios extra arriba y abajo de los cuadros. Éstos deben insertarse en el texto tan cerca del punto de referencia como sea posible. La palabra Cuadro debe colocarse al inicio de la leyenda, usando mayúscula en la primera letra de la palabra (C) y seguida por un punto después del número (arábigo) del Cuadro. La leyenda debe usar el ancho completo de la página impresa y escribirse usando mayúsculas en la primera letra de las palabras principales. Colocar líneas dobles entre la leyenda y el cuadro. Las notas de pie de página de los cuadros deben estar en letras minúsculas y sobrescritas y deben colocarse inmediatamente después del cuadro. Los decimales menores de 1.0 deben ser precedidos por un 0. El autor debe proveer una leyenda en inglés para cada cuadro. Ésta debe ser precedida por la palabra Table, con mayúsculas en la primera letra, e inmediatamente después de la leyenda en español. La revista *Southwestern Entomologist* le puede ayudar con la traducción al inglés de la leyenda de su cuadro(s).
 10. Las gráficas y figuras deben colocarse en el texto tan cerca del punto de referencia como sea posible. Todas las figuras y leyendas deben ser de tamaño legible aun después de una reducción al 25% del tamaño original. La leyenda debe usar el ancho completo de la página impresa. La abreviación Fig., con el número de la figura (número arábigo), debe colocarse al comienzo de la leyenda. Úsese la abreviación Fig. en el texto cuando se haga referencia a una figura. Dejar al menos dos espacios extra arriba y abajo de la figura para que ésta no se confunda con el texto. Si se presenta una fotografía, su descripción debe estar colocada junto a ésta. El autor debe proveer una leyenda en inglés para cada gráfica, figura y fotografía, la cual debe ser precedida por la abreviación Fig., y colocarse inmediatamente después de la leyenda en español. Si se usan fotografías pequeñas, su texto puede ser colocado al lado de la fotografía. La revista *Southwestern Entomologist* le puede ayudar con la traducción al inglés de la leyenda de su figura(s) o fotografía(s).

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Audit Committee Report

Date: July 26, 2022

To: Members of the Society of Southwestern Entomologists

The Audit Committee and I have examined the Society's financial records and the Secretary-Treasurer's Report from January 1, 2021 – December 31, 2021. The financial records for the Society of Southwestern Entomologists are in good order and accurately reported.

Respectfully submitted,
President Society of Southwestern Entomologist



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