Species Diversity and Population Dynamics of Fruit Flies (Diptera: Tephritidae) in Guerrero, Mexico

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Species diversity and population dynamics of fruit flies (Diptera: Tephritidae) in Guerrero, Mexico

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Abstract
Horticulture crops are economically important in the state of Guerrero, México. However, fruit flies (Diptera: Tephritidae) are the most important limiting factor due to the direct damage they cause to fruit crops such as mango (Mangifera indica Lour. [Anacardiaceae]), sweet citrus, guava (Psidium guajava L. [Myrtaceae]), and some Sapotaceae species. The aim of this study was to define fruit fly species diversity and population dynamics in the municipalities of Tetipac and Atoyac de Álvarez, Guerrero. Fruit flies were obtained from the official trapping system of the National Campaign against Fruit Flies (NCFF) of the National Directorate for the Protection Plan, SENASICA, SAGARPA, in the state of Guerrero. Eleven fruit fly species were detected: Anastrepha ludens (Loew), A. striata Schiner, A. obliqua (Macquart), A. serpentina (Wiedemann), A. spotulata Stone, A. bicolor (Stone), A. dentata (Stone), A. chichayoe Greene, Toxotrypana curvicauda Gerstaecker, Rhagoletis ramosae Hernandez-Ortiz, and Zonosemata coccoy Bush (all Diptera: Tephritidae). In Tetipac, we recorded the greatest abundance of fruit flies ($S = 10$) and also the highest values for the Shannon-Wiener ($H'$) diversity index and Simpson ($\lambda$) index ($H' = 1.30; \lambda = 0.68$). Total fruit fly abundance was 1,546 individuals (Tetipac 1,085; Atoyac de Alvarez 461). The main peak populations of fruit flies were recorded from Feb to Oct, coinciding with the phenological stages of fruiting, ripening, and harvesting of fruits of each area. The host range of the most predominant fruit fly species was confirmed by recording flies emerging from fruit. Anastrepha obliqua was detected in mango and jobo (Anacardiaceae) collected in Tetipac and mango from Atoyac de Alvarez. The fruit fly parasitoid Diachasmimorpha longicaudata (Ashmead) (Hymenoptera: Braconidae) was recorded only in Tetipac emerging from fruit flies infesting mango, guava and jobo.

Key Words: Anastrepha spp.; diversity index; Rhagoletis; Toxotrypana; parasitoid

Resumen
La fruticultura es una actividad de importancia económica para el estado de Guerrero; sin embargo, las moscas de la fruta (Diptera: Tephritidae) limitan el desarrollo de esta actividad, ya que causan daños directos a frutales como mango (Mangifera indica Lour. [Anacardiaceae]), cítricos dulces, guayaba (Psidium guajava L. [Myrtaceae]) y algunas sapotáceas. Este estudio se realizó para conocer la diversidad y fluctuación poblacional de moscas de la fruta en los municipios de Tetipac y Atoyac de Álvarez, Guerrero. El material biológico se obtuvo de la red oficial de monitoreo de la Campaña Nacional contra Moscas de la Fruta (CNMF) de la Dirección General de Sanidad Vegetal, SENASICA, SAGARPA en el Estado de Guerrero. Se detectaron 11 especies de moscas de la fruta: Anastrepha ludens (Loew), A. striata Schiner, A. obliqua (Macquart), A. serpentina (Wiedemann), A. spotulata Stone, A. bicolor (Stone), A. dentata (Stone), A. chichayoe Greene, Toxotrypana curvicauda Gerstaecker, Rhagoletis ramosae Hernandez-Ortiz, y Zonosemata coccoy Bush (todas Diptera: Tephritidae). En Tetipac se registró el mayor número de especies ($S=10$) y de acuerdo a los índices Shannon-Wiener ($H'$) y Simpson ($\lambda$) mayor diversidad ($H' = 1.30; \lambda = 0.68$). La abundancia total fue de 1,546 ejemplares (Tetipac 1,085 y Atoyac de Alvarez 461). Los principales picos poblacionales de moscas de la fruta se presentaron de febrero a octubre, coincidiendo con las etapas fenológicas de fructificación, maduración y cosecha de frutos de cada área. El rango de especies de hospederos de moscas de la fruta más predominante fue confirmado al registrar las moscas que emergieron de fruta muestreada. Anastrepha obliqua fue detectada en mango y jobo (Anacardiaceae) colectados en Tetipac y de mango en Atoyac de Alvarez. El parasitoide de moscas de la fruta Diachasmimorpha longicaudata (Ashmead) (Hymenoptera: Braconidae) fue registrado solo en Tetipac de frutos hospedantes de moscas de la fruta como mango, guayaba y jobo.

Palabras Clave: Anastrepha spp.; índice de diversidad; Rhagoletis; Toxotrypana; parasitoid

Tropical horticulture is economically important in Mexico; in the last few years Mexico has become one of the main exporters of mango (Mangifera indica Lour. [Anacardiaceae]) and papaya Carica papaya L. (Caricaceae) (Villegas-Monter & Mora-Aguilera 2011). Of all the states in Mexico, Guerrero produces the most mangoes, particularly the cultivars Manilla and Atauilo (SIAP 2016).

A key threat to horticultural development in Guerrero, and in other states, is the complex of phytophagous pests, amongst which some species in the family Tephritidae (fruit flies) are the most important (Aluja 1993, 1994; Hernández-Ortiz et al. 2010). Worldwide there are nearly 4,000 species belonging to 500 genera in the Tephritidae (White & Elson-Harris 1992), and almost 200 species are considered important pests due to the direct losses that they cause to many fruit tree species (Norrbom et al. 2012). Globally, the most widespread genera of fruit flies are Bactrocera, Ceratitis, Rhagoletis, Toxotrypana, and Anastrepha (all Diptera: Tephritidae) (Hernández-Ortiz et al. 2010). Anastrepha

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is one of the most diverse genera and is endemic to the Americas (Aluja 1994; Norrbom et al. 1999; Norrbom & Korytkowski 2009, 2011). The genus has 209 described species (Hernández-Ortiz et al. 2010), 7 of which are considered economically important: *Anastrepha ludens* (Loew) (Mexican fruit fly); *Anastrepha obliqua* (Macquart) (West Indian fruit fly); *Anastrepha fraterculus* (Wiedemann) (South American fruit fly); *Anastrepha suspensa* (Loew) (Caribbean fruit fly); *Anastrepha serpentina* (Wiedemann) (sapote fruit fly); *Anastrepha striata* Schiner (guava fruit fly); and *Anastrepha grandis* (Macquart) (cucurbit fruit fly) (all Diptera: Tephritidae) (Hernández-Ortiz et al. 2010). Mexico has 37 species of *Anastrepha*; and the highest species diversity is found in the states of Chiapas and Veracruz (25 and 24 species, respectively). In Guerrero, 12 species have been recorded: *Anastrepha tripunctata* Wulp, *Anastrepha bicolor* (Stone), *Anastrepha pallens* Cockerillet, *Anastrepha montei* Lima, *Anastrepha spatulata* Stone, *Anastrepha chichlaiay* Greene, *A. serpentina* (Wiedemann), *A. striata* Schiner, *Anastrepha acris* Stone, *Anastrepha distincta* Greene, *A. ludens* (Loew), and *A. obliqua* (Macquart) (all Diptera: Tephritidae) (Hernández-Ortiz 2007).

Currently, the National Campaign against Fruit Flies (NCFF) is active in 26 states of Mexico, where monitoring activity (adult trapping and fruit sampling) and control strategies are implemented under an Integrated Pest Management scheme, with the aim of establishing and keeping fruit fly-free areas and reducing fruit fly populations to low prevalence levels (= fruit flies per trap per d [FTD]) of ≤ 0.01) (SENASICA 2017). It is recognized that understanding the population dynamics of these pests in particular regions of the country can inform control measures (Aluja 1993). The objectives of this research were to: (a) update knowledge on the diversity of *tephritid* fruit flies from the family Tephritidae and determine the diversity and seasonal distribution of *tephritid* fruit flies in the state of Guerrero; and (b) identify fruit fly hosts in the municipalities of Tetipac and Atoyac de Álvarez, Guerrero, Mexico.

Materials and Methods

**STUDY AREA**

Two municipalities in Guerrero, where the NCFF carries out control actions, were selected: (1) Tetipac, and (2) Atoyac de Álvarez. Tetipac is located in the north of the state (18.3544°N, 99.3247°W) (INEGI 2009a, b) at an elevation of 1,660 meters above sea level (masl). It has a temperate sub-humid climate with rains during the summer and an annual precipitation of 800 to 1,300 mm (INEGI 2016). Tetipac produces more guava cv. ‘Media China’ than any other municipality in the state over an area of 157 ha with an annual yield of 2,179 tonnes (SIAP 2016). Atoyac de Álvarez is located in the south of the state (17.0434°N, 100.0632°W) at an altitude of 40 masl, and it has a warm sub-humid climate with rains during the summer and an annual precipitation of 1,100 to 2,500 mm (INEGI 2016). Atoyac de Álvarez has the second highest production of mango cv. ‘Ataúlfo’ of any other municipality in the country over an area of 1,336 ha, with an annual yield of 18,028 tonnes (SIAP 2016).

**ADULT SAMPLING**

The official monitoring network (trapping) of the NCFF was used to capture adult fruit flies. We analyzed the material collected from 82 traps in Tetipac and 496 traps in Atoyac de Álvarez. The traps used were ‘Wet’ Multilure International traps (Better World Mfg. Inc., Fresno, California, USA), baited with 3 torula yeast tablets (A Better Trap Inc. [torula yeast and borax pellets], Fresno, California, USA) diluted in 250 mL of water. This type of trap is used especially for monitoring species of fruit flies (Thomas et al. 2001; García-Ramírez et al. 2008). Traps were checked and the bait replaced every 7 d by technical personnel from the Plant Health State Committee of Guerrero (CESAVEGRO) from 19 Oct 2015 to 17 Oct 2016. Adult fruit flies from the traps were placed in 250 mL glass jars filled with 70% ethanol prior to identification.

**FRUIT SAMPLING**

To identify fruit fly hosts, samples of fruit were taken from trees and from those that had fallen to the ground; in both cases fruits that had clear damage due to fruit fly larvae were randomly collected during the fruit production season (May to Sep) (Silva et al. 2010; Nicáio & Uchôa 2011). The number of collection sites varied because the objective of this study was to define host diversity not levels of damage, but were selected from those sites where the traps were established. Collected fruits included mango cv. ‘Criollo’ (Manihot indica L.), guava cv. ‘Media China’ (Psidium guajava L.), jobo (Spondias purpurea L) and bonete or cuaguayote (Jacaranda mexicana L. [Caricaceae]). Collected fruits were placed in 28 × 18 × 11 cm plastic containers on a substrate of Agrolita (Agrolita de Mexico, S.A. de C.V. Tlánepanitla de Baz, Estado de México) for development of pupae from any larvae emerging from the fruit. Each container was covered with organza to allow ventilation and to capture any emerging adult fruit flies and parasitoids. All emerging adult fruit flies and parasitoids were stored in 70% ethanol prior to identification.

**TAXONOMY**

Adult fruit flies were identified using the dichotomous keys of Hernández-Ortiz (1985), White & Elson-Harris (1992), Aluja (1993), Norrbom (2002), Hernández-Ortiz et al. (2010), and the interactive keys of Norrbom et al. (2012). Adult parasitoids were identified by comparing them with voucher specimens provided by Dr. J. Refugio Lomeli. Voucher specimens were deposited in the Insect Collection of the Colegio de Postgraduados, Montecillo, State of Mexico, Mexico (CPEAM).

**STATISTICAL ANALYSES**

To measure species diversity of Tephritidae, data from fruit flies captured on Multilure traps were analyzed, and the Shannon-Wiener (H’) equitability index was used, which expresses the species diversity within a community and the uniformity of the important values taking into account all the species in a sample following the equation $H' = \sum p_i \ln p_i$; where $p_i$ represents the proportional abundance of species $i$ (Moreno 2001). Also, the Simpson dominance index ($\lambda$) was used, which calculates the probability that 2 individuals selected at random from a sample will belong to the same species, through the equation $\lambda = \frac{\sum p_i}{p_i^2}$ (Simpson 1949). This index is strongly influenced by the importance of the most dominant species, and its value is inverse to the equitability, and diversity is calculated as $1-\lambda$. The statistical software InfoStat version 2008 was used for all statistical analyses (Balzarini et al. 2008).

To determine the behavior of the population of each species of fruit fly and its plant health category according to the NCFF, the fruit flies per trap per d (FTD) index was employed, using weekly abundance data, following the equation $F/T*D$, where $F$ represents the number of fruit flies trapped in the exposure period of the trap, $T$ the number of traps inspected in the period, and $D$ the number of d that the traps were exposed in the field. The NCFF have established 3 Plant Protection categories: Free Fruit Fly zone, FTD = 0.0; Low Prevalence Zone, FTD ≤ 0.03; and Zone Under Plant Protection Actions, FTD > 0.01. Data for fruit flies trapped were analyzed to obtain averages using descriptive statistics from Excel (Microsoft Office, ver. 2010®).
Results

DIVERSITY OF FRUIT FLIES FROM TRAPS

Eleven species of Anastrepha, Rhagoletis, Toxotrypana, and Zonosemata were recorded. In Tetipac, 10 species were recorded, and in Atoyac de Álvarez only 5 (Table 1). Shannon-Wiener ($H^\prime$) and Simpson ($\lambda$) indices show that the diversity of fruit fly species was higher in the municipality of Tetipac than the municipality of Atoyac de Álvarez ($H^\prime = 1.30; \lambda = 0.68$) (Table 2).

A total of 1,546 fruit flies were captured between Oct 2015 and Oct 2016. In the municipality of Tetipac, a total of 1,085 individuals were recorded (647 females, 438 males). Anastrepha ludens was the most abundant species accounting for 41.29% of all flies captured, followed by A. striata (32.44%), and A. obliqua (20.92%). The remaining 7 species of fruit fly each were present at a level of less than 3% of the population.

In the municipality of Atoyac de Álvarez, a total of 461 individuals were recorded (353 females, 108 males). Anastrepha obliqua was the most abundant species accounting for 90.67% of all flies captured, followed by A. striata (6.7%). The remaining species each were present at a level of less than 2% of the population (Table 1).

Fruit flies per trap per day analysis of the number of fruit flies trapped in Tetipac showed a relatively constant population of A. ludens between Feb and Oct 2016 (Fig. 1). Population peaks of A. striata occurred in Nov 2015, Mar 2016, and between Aug and Oct 2016. Population peaks of A. obliqua first occurred in Nov 2015 and between Aug and Oct 2016. Populations of the remaining species were low and recorded only in 2016. For example, A. serpentina occurred in Jun and Aug, A. spatulata in Mar and Apr, A. bicolor from Apr to Jul and in Sept, A. dentata in Jul and Sep, Toxotrypana curvicauda in Jun and Jul, Zonosemata cocomoyoc in Jul and Sep, and Rhagoletis ramosae in Sep (Fig. 1).

In Atoyac de Álvarez, A. obliqua was recorded in 46 of the 53 wk monitored, and it was abundant between Feb and Sep 2016 (Fig. 2). The populations of the remaining species were low, and their presence was sporadic. For example, A. striata was recorded in Dec 2015, between Feb and May 2016, and between Sep and Oct 2016; A. ludens occurred in Mar and May 2016; A. chiclayae was recorded only in May 2016; and T. curvicauda was recorded in Nov 2015 (Fig. 2).

DIVERSITY OF FRUIT FLIES FROM FRUIT SAMPLES

The most important population peaks of fruit flies in both municipalities was related to the phenological stages of the fruit trees in each study area, such as fruit development stage, fruit maturity, and harvesting period (Fig. 3).

The species of fruit flies detected by direct fruit sampling from the main host species in Tetipac were: A. ludens in mango; A. striata in guava; A. obliqua in jobo and mango; and T. curvicauda in bonete. In Atoyac de Álvarez only A. obliqua was recorded in mango. In addition to the emergence of adult fruit flies sampled from fruits on trees and fallen fruits, the emergence of adults of the parasitoid D. longicaudata was recorded from mango, guava, and jobo in Tetipac; no parasitoids were recorded emerging from fruit collected in Atoyac de Álvarez (Table 3).

Discussion

In Mexico, research on the population fluctuations of Tephritidae have focused mainly on species of economic importance (González-Hernández & Tejada 1979; Thomas 2003; Tucuch-Cauich et al. 2008; Aluja et al. 2012; Vanoye-Eligio et al. 2015; Hernández-Adame et al. 2015). However, some studies have provided full lists of the species of fruit fly present in a given area, their primary hosts, and at least some data on their population dynamics (Celedonio-Hurtado et al. 1995; Miranda-Salcedo & Leyva 1996; Martínez & Serna 2005; Martínez-Alava 2007; Nolasco & Lannaccone 2008; Gonzáles et al. 2011; Miranda-Salcedo 2012; René-Arias et al. 2014; Montoya-Álvarez et al. 2014; Vanoye-Eligio et al. 2014).

Of the 11 species of fruit flies previously recorded in the state of Guerrero, 5 also were found in this study and in both municipalities, and they are all considered of economic importance. These are A. ludens, A. striata, A. obliqua, A. serpentina, and T. curvicauda (Aluja 1993; Hernández-Ortiz & Aluja 1993; Hernández-Ortiz et al. 2010). In the municipality of Tetipac, in addition to these species, A. bicolor, A. spatulata, and A. dentata also were found, which have already been documented in Mexico (Hernández-Ortiz 2007), as well as A. ramosae, which was described for the first time from material from Guerrero and Michoacán (Hernández-Ortiz 1985), and Z. cocomoyoc, a species that had been recorded already in the states of Guerrero, Chiapas, and Morelos (Norrbom 2002).

The fact that higher species richness and diversity of fruit fly species were recorded in Tetipac ($S = 10$ and $H^\prime = 1.30, \lambda = 0.68$) than in Atoyac de Álvarez ($S = 5$ and $H^\prime = 0.39, \lambda = 0.17$) is probably due to the presence in every backyard in Tetipac of a greater diversity of backyard fruit trees, such as jobo, orange [Citrus sinensis L. [Rutaceae], mango, papaya, mamey (or sapote) [Pouteria sapota Jacq. [Sapotaceae]], and peach [Prunus persica L. [Rosaceae]] in addition to guava, which is the main crop of the region. According to Aluja (1993) and Hernández-Ortiz (2007), backyard fruit trees are hosts of a range of fruit fly species. In contrast, Atoyac de Álvarez is an area where commercial mango production predominates due to its great economic importance; mango also is grown in every backyard, and is found amongst wild vegetation. The sites where NCFF traps were established did not allow the detection of a higher diversity of species owing to the reduced number of alternative hosts near commercial orchards.
During the period from Oct 2015 to Oct 2016, the total abundance of fruit flies was higher in Tetipac (70.18%) than in Atoyac de Álvarez, and the most common species was A. ludens, which is characterized by a higher capacity for adaptation to more elevated areas (> 900 masl) (Hernández-Ortiz 1992), followed by A. striata and A. obliqua. These 3 species were present throughout the entire monitoring period, because the season for fruit production of each tree species occurs in different months; thus, the insects have host fruits available throughout the year. This confirms other studies where fruit production has been documented as the main factor affecting fruit fly populations in tropical environments (Celedonio-Hurtado et al. 1995; Nolasco & Lanacone 2008; Tucuch-Cauich et al. 2008; González et al. 2011; Aluja et al. 2012; Hernández-Adame et al. 2015; Vanoye-Eligio et al. 2015). Populations of the other fruit fly species detected in this study and others occurred sporadically from Mar to Sep 2016 with only a few individuals captured.

During the period from Oct 2015 to Oct 2016, the total abundance of fruit flies was lower in Atoyac de Álvarez (29.8%) than in Tetipac (70.2%), and the most common species was A. obliqua, which is more adapted to areas at a low elevation (< 500 masl) (Hernández-Ortiz 1992); the species was recorded in 46 of the 53 wk monitored, and from Feb to Sep 2016. In contrast, in this municipality, populations of

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**Fig. 1.** Fruit flies per trap per day of fruit flies in Tetipac, Guerrero 2015–2016. Other = A. serpentina, A. spatulata, A. bicolor, A. dentata, T. curvicauda, Z. coccoc, and R. ramosae.

**Fig. 2.** Fruit flies per trap per day of fruit flies in Atoyac de Álvarez, Guerrero 2015–2016. Other = A. chiclayae and T. curvicauda.
A. striata, A. ludens, A. chilcayae, and T. curvicauda were small; only 1 individual of T. curvicauda was recorded in the entire year.

In both areas of study, the populations of fruit flies collected were predominantly female, as seen in other studies (Nolasco & Lannacone 2008; Espolador-São João et al. 2014; René-Arias et al. 2014; Montoya-Alvarez et al. 2014; Hernández-Adame et al. 2015) (Table 2). This is because the food bait used in traps to catch adults (torula yeast and borax) is based on proteins that are important for oocyte formation in females.

The presence of the most common species of fruit flies throughout the study, such as A. ludens, A. striata, and A. obliqua, coincided with the phenological stages of fruit production, fruit maturity, and harvest period for the tree species present in each region (Fig. 3). Collecting infested fruits helped to confirm the host range and host preference of the fruit fly species associated with the municipalities of Tetipac and Atoyac de Álvarez. In Tetipac, A. ludens was recorded in mango, A. striata in guava, A. obliqua in jobo and mango, and T. curvicauda in bonete, as has been recorded previously (Aluja 1993, 1994; Hernández-Ortiz & Aluja 1993; Hernández-Ortiz 2007; Martínez-Barrera et al. 2015). In Atoyac de Álvarez, A. obliqua was recorded only in mango (Table 3).

Analysis of the fruit flies per trap per d data for 2015–2016 for the economically important fruit fly species showed that in Tetipac, A. ludens, A. striata, and A. obliqua were the species most frequently captured in the area. Population peaks in most cases had fruit flies per trap per d values higher than 0.01 (zones under plant protection actions). This means that the NCFF must apply several control strategies, such as chemical, cultural, and biological control (IPM), if fruit fly populations are to be controlled. These 3 fruit fly species may be present at the range of altitudes of the sites where the study was conducted (50–1,660 masl), but this depends on the host tree diversity. In contrast, in Atoyac de Álvarez, A. obliqua was the species most frequently captured, while populations of A. striata, and A. ludens were small, and the population peaks always had fruit flies per trap per d values below 0.01 (low prevalence zone). For this category it is sufficient for the NCFF to release sterile flies to suppress fruit fly populations. In this area, altitude may not influence host tree diversity for fruit flies, because mango trees are present more frequently.

Finally, of the 4 fruit fly species of economic importance detected in the 2 study sites, A. serpentina had the smallest populations in Tetipac, with fruit flies per trap per d values always below 0.01, and in Atoyac de Álvarez no individuals of this species were caught in traps. According to the Mexican Plant Protection Standard NOM-023-FITO-1995 (SAGAR 1999), the incidence levels of these 4 fruit fly species are used to define the plant protection zones or categories that have been established for the NCFF, namely Free Zone with FTD = 0.00; Zone of Low Prevalence (with FTD ≤ 0.01; and Zones under Plant Protection Actions (FTD > 0.01, with an upper limit of > 0.08); management strategies proposed by the NCFF are carried out taking these categories into account.

In general, it can be observed that fruit fly populations in Tetipac maintain fruit flies per trap per d values of > 0.01, whereas in Atoyac, populations maintain fruit flies per trap per d values of < 0.01. The results of the present study show that the municipality of Tetipac is in the Zones under Plant Protection Actions category, which means it has high levels of fruit fly incidence. In contrast, Atoyac de Álvarez is in the Zones of Low Prevalence category, and it may attain the Free Zone category in the future, which would reduce management costs, and producers could freely trade fruits such as mango in domestic and international markets without quarantine restrictions.

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