Influence of the height of multilure traps in the collection of Ceratitis capitata (Wiedemann) (Diptera: Tephritidae)

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Abstract. The Mediterranean fruit fly Ceratitis capitata (Wiedemann) is a polyphagous pest adapted to tropical and subtropical climates, which are responsible for the highest share of fruit production. Fruit fly surveys are frequently done by installing traps at heights easily reached by the collector, between 1.5 and 2.5 meters. This study aims to inform fruit fly monitoring strategies by assessing two trap heights (around 2 m and 10 m) in four environments (agricultural and forest) at the “Luiz de Queiroz” Campus, Piracicaba, São Paulo state, Brazil. Each collection environment was considered a block, and eight multilure traps were installed at each of the two heights on four plants per block. A generalized linear model was used for non-normal data with negative binomial distribution to compare the abundance of Ceratitis capitata between trap heights and areas. The higher traps, positioned at around 10 m, collected significantly more specimens of C. capitata than those at approximately 2 m in height. The Mediterranean fruit fly was more frequent in the cultivated environments and areas with human activity than in natural areas.

Keywords: Collection; Mediterranean fruit fly; population fluctuation.

Fruit flies (Tephritidae) are the most economically important group for fruit farming worldwide (Norrbom et al. 1999; Zucchi 2000; Aluja & Mangan 2008; Savaris et al. 2016; Almeida et al. 2019; Zida et al. 2020). In the Americas, the most economically important species belong to the genera Anastrepha and Rhagoletis, as well as the introduced species Ceratitis capitata (Wiedemann) and Bactrocera carambolae Drew & Hancock (Norrbom et al. 1999, 2018; Zucchi 2000; Minhibo et al. 2018).

The Mediterranean fruit fly C. capitata was detected in Brazil in 1901 (Herbing 1901; Zucchi 2015). It is a polyphagous species that has been recorded in 97 host plants in Brazil, with a particular affinity for species of the families Myrtaceae, Rutaceae, Rosaceae, Anacardiaceae, and Sapotaceae (Zucchi & Moraes 2012). Although its initial dispersion occurred in the country’s southeastern and southern regions, C. capitata has also been recorded in the northern (Ronchi-Teles & Silva 1996), northeastern (Morgante 1991), and central-western (França et al. 1995) regions of the country. The only states that have no records of the species are Amazonas and Amapá (Zucchi & Moraes 2012; Zucchi 2015).

The Mediterranean fruit fly has been one of the main phytosanitary problems for several fruit trees cultivated in Brazil, representing one of the main obstacles for the expansion of fruit farming and exports of fresh fruits (Costa et al. 2011; Zucchi 2015). Intensive monitoring of this species has therefore been performed in several states in an effort to detect its presence and infestation levels (Malawski et al. 1980; Silva et al. 2011).

Monitoring of C. capitata has commonly been performed with traps arranged at heights ranging from 1.5 to 2.5 m, where they can easily be reached by the collector or placed in the crown of fruit trees (Montes & Raga 2006; Paranhos 2008). However, few studies involve traps for this species positioned at greater heights, which may offer additional insights for managing this species, especially in areas with large fruit trees.

Given the paucity of studies on the collection of these flies at higher heights, this study aims to evaluate the effectiveness of multilure traps installed at heights of 2 and 10 m capturing C. capitata in agricultural and forestry environments at the “Luiz de Queiroz” Campus, Piracicaba, São Paulo state, Brazil.

MATERIAL AND METHODS

Study area. The study was conducted at the “Luiz de Queiroz” Campus of the “Luiz de Queiroz” College of Agriculture, University of São Paulo (ESALQ/USP), Piracicaba, São Paulo, from April 2019 to March 2020 (Figure 1). The vegetation present on the Campus belongs to the Atlantic Forest biome, more specifically the Semideciduous Seasonal Forest type, which is characterized by emerging plants with an irregular canopy between 15 and 20 m high, which lose their leaves in the dry winter (Lima 2000; Alexandrino et al. 2013). The main plant families present on the Campus are Anacardiaceae, Apocynaceae, Bombacaceae, Fabaceae, Lauraceae, Lecythidaceae, and Myrtaceae (Rodrigues 1999). The climate of the study area, according to Köppen classification, is the Cfa type, with a dry winter and rainy and hot summer, and average annual rainfall of between 1,300 and 1,600 mm (Alvarens et al. 2013). However, the region has recently experienced conditions more similar to the tropical Aw climate, characterized by dry winters with an average annual rainfall of 1,100 to 1,300 mm (Alvarens et al. 2013; Dias et al. 2017).
The collection sites of *C. capitata* have contrasting environments in terms of their vegetation, as follows: the Areão farm (Figure 2A), which has cultivation areas of acerola cherry (*Malpighia emarginata* DC.), banana (*Musa* sp.), coffee (*Coffea arabica* L.), jocote (*Spondias purpurea* L.), mango (*Mangifera indica* L.), loquat (*Eriobotrya japonica* (Thunb.) Lindl.), and ‘uvaia’ (*Eugenia pyriformis* Cambess.), in addition to a mix of native and exotic tree species. The Central Area (Figure 2B) and the Piracicamirim River (Figure 2C) have native and exotic tree plants forming a diffuse mosaic. Finally, Monte Olimpo (Figure 2D) is an area with a native forest and little human activity.
Collection. We installed 32 multilure traps on the "Luiz de Queiroz" Campus, seeking to sample different environments to collect specimens of the Mediterranean fruit fly (Figure 1). The traps were distributed in four blocks with eight traps each and were installed on four different plants in each block in the agricultural and native forest areas. The blocks were in the Areão Farm (22°41’41.90" S 47°38’25.22" W, 556 m), Central Area (22°42’40.56" S 47°37’55.71" W, 560 m), Piracicamirim River (22°42’22.58" S 47°37’39.05" W, 531 m), and Monte Olimpo (22°43’9.41" S 47°36’43.37" W, 585 m). In the study areas, the traps were arranged as follows: a high trap at a height of approximately 10 m (Figure 3A, indicated by the red arrow) and a low trap at a height of approximately 2 m (Figure 3C), both on the same plant with a minimum distance of 100 m between pairs of traps in a single block, forming four collection stations per block. The high traps were installed with slings and nylon lines to reach the plants' branches in a leafy canopy, which corresponds to approximately 10 m in height.

The high traps were suspended in the trees by nylon ropes with a diameter of 3 mm, which were lowered during the surveys to remove the Mediterranean fruit fly (Figure 3A-B). The low traps were installed with extension cables, preferably in the same plants in which the high traps had been installed (Figure 3C). To locate the traps in each collection station, within each block, colored tape (Figure 3C) was used, with the block data and trap number written on it.

As an attractor for collecting the Mediterranean fruit fly, a product made from putrescine and ammonium acetate was used, consistent with common practice in monitoring programs for fruit flies of economic interest, such as species of Anastrepha (Thomas et al. 2001).

In each trap, 250 mL of propylene glycol (C₅H₈O₂) was placed to preserve the collected insects. The traps were reviewed every 15 days, and the collected material was stored in properly labeled plastic bottles containing propylene glycol or 99% ethanol. Since C. capitata is the only species of the genus Ceratitis recorded in Brazil, the specimens' identification was based exclusively on external morphological characters.

Voucher specimens were deposited in the collection of the "Luiz de Queiroz" Entomology Museum (MELQ), Department of Entomology and Acralogy, of the "Luiz de Queiroz" College of Agriculture, University of São Paulo (ESALQ/USP). This study was supported by the project "Integrative Taxonomy of Brazilian species of the genus Anastrepha (Diptera: Tephritidae) based on the study of larvae and adults". The collections were made with the authorization of the Brazilian Biodiversity Authorization and Information System (SISBIO) under number 61549.

The map of the collection areas was made in Quantum GiS 2.8 software (QGIS 2020). Map coordinates were obtained at each collection point using a Garmin 78S GPS.

The abundance of C. capitata across areas and sites was evaluated using a half-normal probability plot with simulation envelope generated with the hnp package (Moran et al. 2017). The abundance of C. capitata across areas and sites was compared with an analysis of variance (ANOVA) using results associated with the F-Test. Based on the ANOVA results, the means were compared through the confidence interval (95%) of the linear predictor of the adjusted model.

RESULTS AND DISCUSSION

One hundred specimens of C. capitata were collected at the "Luiz de Queiroz" Campus, of which 57 were females and 43 were males. Seventy-eight specimens were captured in the high traps and 22 in the low traps (Table 1).

There was a significant difference in the number of C. capitata specimens captured at the 2 and 10 m heights, with a higher number of specimens captured in the tall traps at all sites except for the forest environment at Monte Olimpo (Table 1). This result is consistent with the preference of C. capitata for cultivated, anthropic environments over native forest environments. Canal et al. (1998) and Souza et al. (2008) observed that, despite being able to live in a wide range of hosts, the Mediterranean fruit fly prefers environments and hosts in urban and anthropized open areas.
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The difference between the average abundance catches at each sampling site was significant between Areão Farm and Monte Olimpo, with more specimens collected at Areão Farm. The other areas were statistically similar in terms of the total number of fruit flies collected (Figure 4).

**Table 1.** Number of specimens of *Ceratitis capitata* collected with multilure traps in four collection sites at the “Luiz de Queiroz” Campus, Piracicaba, state of São Paulo, Brazil, between April 2019 and March 2020.

| Areas             | Males | Females | Heights (≤ 2 m) | Heights (≥ 10 m) | Total |
|-------------------|-------|---------|----------------|------------------|-------|
| Areão Farm        | 32    | 39      | 21             | 50               | 71    |
| Central Area      | 4     | 11      | 0              | 15               | 15    |
| Piracicamirim River | 7    | 6       | 0              | 13               | 13    |
| Monte Olimpo      | 0     | 1       | 1              | 0                | 1     |
| **Total**         | **43**| **57**  | **22**         | **78**           | **100**|

High traps collected more specimens of *C. capitata* in the human-altered environments or those surrounded by such areas (Central Area, Areão Farm, and Piracicamirim River; Table 1).

On the other hand, in the more natural forest environment (Monte Olimpo), only one female was collected in a low trap installed near the forest edge, meaning that this data could not be compared with the other sampled areas due to its low variability (Figure 5). This result may be due to the preference of *C. capitata* for open and human-altered areas (Canal et al. 1998; Vera et al. 2002; Souza et al. 2008).

The low number of *C. capitata* may have been influenced by the smaller number of its hosts at the Monte Olimpo study area and the low effectiveness of the lure used. However, it should be noted that even in areas with coffee trees (Areão Farm), more specimens of *C. capitata* were collected in the tall traps (installed in the canopy of the inga trees - *Inga* sp.) than in the low ones (coffee trees), although *C. capitata* prefers exotic hosts such as coffee to native ones such as the inga trees (Malavasi & Morgante 1980; Araújo et al. 2005).

At Areão Farm, it was thus surprising that fewer *C. capitata* individuals were collected in the low traps placed among coffee plants (primary host of the Mediterranean fruit fly) (*Puzzi & Orlando 1965; Montes et al. 2012*) than in the high traps installed in inga trees. However, given the low total number of flies collected and the short collection period, this observation requires additional verification, ideally including the phenology of the host plants. The collected fruit flies at different heights proved to be feasible for these objectives and should be considered in monitoring strategies for this pest in fruit cultivation. The height at which *C. capitata* in Latin America predominantly flies is still unknown and thus requires further studies. Interestingly, two studies have been conducted on the vertical distribution of *Anastrepha striata* Schiner in Costa Rica. In the Costa Rican guava (*Psidium friderichi-talinum* (O.Berg) Nied.) orchards, more specimens were collected at heights between 6 and 10 m in Puerto Viejo de Sarapiquí (Hedström & González 1987) and at heights of 7 and 10 m in Guacimo (Hedström 1992). These authors suggested that more fruit flies were collected at 7 and 10 m than at 4 or 14 m because of the number of guava fruits at these heights. In addition, more flies were collected at the height of 8 m than at 4, 6, and 10 m in the guava trees.

There was no significant difference in the occurrence of Mediterranean fruit flies between months of the year, though their presence was greater during the winter period of May to August (Figure 6).

It can be inferred that the occurrence of *C. capitata* in the study sites was similar throughout the year, probably due to...
the seasonality of fruiting of the different native and exotic plant species and the potential hosts of the Mediterranean fruit fly in the study area.

In months with lower mean temperatures, the number of C. capitata individuals was significantly higher. The highest population fluctuation occurred from May to August (Figure 6), which coincides with the fruiting and post-fruiting period of the coffee trees in the Área Farm area, where 71% of Mediterranean fruit fly specimens were collected. *Ceratitis capitata* uses the fruits of the coffee tree as hosts for the development of its larvae, which may cause damage to the crop (Torres et al., 2010).

The Mediterranean fruit fly occurs mainly in human-altered environments and is less common in dense and preserved forest environments (Canal et al. 1998; Vera et al. 2002). It has been associated with 97 native and exotic host species in Brazil (Zucchi & Moraes, 2012), meaning that its year-round presence is likely due to constant availability of hosts on the Campus produce fruits successively throughout the year (succession of hosts), ensuring the continued presence of *C. capitata* throughout the study area.

Since more *C. capitata* individuals were captured at a height of 10 m than at 2 m, management and control efforts in commercial fruit cultivation should focus on higher parts of the canopy. Monitoring traps located higher in the canopy would be more effective at collecting the pest. However, this observation needs to be tested in commercial orchards and different species of fruit trees.

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