Population Fluctuation of Fruit Flies (Diptera: Tephritidae) in Domestic Orchards in the Paraíba Swamp Region, Brazil

ABSTRACT

Knowledge about the population fluctuation of fruit flies (Diptera: Tephritidae) is an important tool for control methods adoption as recommended by Integrated Pest Management (IPM). The objective of the research was to evaluate the fruit flies population fluctuation in eight domestic orchards from Paraíba swamp region and to correlate this information with the plant phenology as well as the meteorological elements. This research was developed in two rural properties of each municipality, georeferenced, identified according to fruit species diversity criterion. Adult fruit flies capture were realized by plastic traps containing 300 ~~mL~~ ml of 5.0% hydrolyzed protein aqueous solution (Bio Anastrepha®) as food attractant. Climatic data were obtained daily by thermo hygrometers and through the Executive Agency of Water Management of the State of Paraíba - AESA. During the study period, a total of 3.159 fruit flies were collected, with 10 species belonging to the genus Anastrepha and *Ceratitis* capitata ~~species~~. Fruit flies infestation was observed monthly for both genera, *Anastrepha* and *Ceratitis*, however, this first stood out in the whole year. The MAD index in a few municipalities was superior to 0.5 being recommended to do fruit flies control in these areas. The seasonal occurrences, as well as the ~~*Tephritidae*~~ Tephritidae infestation rates in the domestic orchards are related to the availability of fruits.

Keywords: *Seasonal occurrence; MAD index*; *Tephritidae*; *Weather Element*

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1. INTRODUCTION

In Brazil, fruit production occurs in all regions, but it predominates in tropical and subtropical climates, where phytosanitary problems represent one of the main obstacles to the expansion of fruit production. The outlook for the sector is optimistic, therefore it is necessary to develop strategies to control pests that compromise production and/or exports [1].

Among the pests, which can be defined as any species, race or plants biotype, animals or pathogen vectors, fruit flies are prominent because of their destructive potential. These insects are considered to be the biggest fruticulture pest in the world, it is easily adaptable when introduced in another region and, in many cases, can compromise up to 100.0% of production [2]. Knowledge about the population fluctuation and the time of greatest occurrence of a certain fruit flies species economically important are indispensable requirements in order to establish an efficient and rational control since they allow being feasible the planning of effective management strategies [3].

Several factors, such as climate, altitude, geographic location, phenology and host succession (primary or secondary), may influence the abundance of certain species of fruit flies throughout the year [14]. An example is the population explosions of some species of *Anastrepha* at certain times of the year, with peaks of the density of adult individuals directly related to hosting phenology and their high reproductive potential, while other species, such as *A. fraterculus*, do not develop at temperatures below 10 ºC and above 35 ºC [5].

Traps usage allows to verify these insects population fluctuation and to relate them to the abiotic factors, mainly those associated to the climate, helping to define the seasons of the greater or lesser probability of infestations [6]. Despite the growing advent of tephritid research in the country over the years, information about the bioecology of fruit flies is still scarce in several regions [7]. Thus, the objective of this study was to evaluate the population fluctuation of fruit flies in the Paraíba swamp region and to correlate this information with plant phenology and meteorological elements.

2. MATERIAL AND METHODS

The study area is located in the Mesoregion of the Agreste of Paraíba and Microregion of the swamp of Paraíba including the municipalities of: Alagoa Grande (S 07° 01 '53.6 "W 035° 38' 12.1"), Alagoa Nova (S 07° 04' 56.3 "W 035° 48' 53.1"), Areia (S 06° 59' 22.7 "W 035° 44' 00.2"), Bananeiras (S 06° 43' 44.3 "W 035° 39' 24.0"), Borborema (S 07° 06' 40.0 "W 035° 49' 10.5"), Pilões (S 06° 56' 45.4 "W 035° 39' 38.2") and Serraria (S 06° 49' 03.8 "W 035° 39' 19.4"). The research was developed in two rural properties of each municipality, georeferenced, identified according to fruit species diversity criterion. Population survey was carried out from July 2015 to June 2016.

Adult fruit flies were obtained with the aid of plastic traps (PET), two traps/plants were installed in each sampling areas. The traps were placed at the mid-height of the tree canopy and contained 300 mL of an aqueous solution of hydrolyzed protein up 5.0% (Bio Anastrepha®) as food attractant.

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The flasks were inspected biweekly, the occasion that captured fly specimens were collected and food attractants were replaced. These specimens were washed with water in a sieve and then packed in plastic containers with 70.0% hydrated alcohol, properly labeled and then sent to the Laboratory of Invertebrate Zoology of the Agricultural Sciences Center of the Federal University of Paraíba - Areia/PB, where the males and females of the *Anastrepha* and *Ceratitis* genuses were screened and stored in 70.0% hydrated alcohol for later species identification.

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The specimens of fruit flies were separated by sex and only the females were identified through the aculeus of the ovipositor, since the males did not present the diagnostic characters for the specific identification [8], using identification keys [9]. The females collected from the *Anastrepha* genus were identified by Dra. Clarice Diniz Alvarenga Corsato - State University of Montes Claros, Janaúba, MG.

Anastrepha and *Ceratitis* species surveys for this study of population fluctuations were carried out through individuals collected in the PET traps, during a period of one year, the necessary time to carry out the species population fluctuation in the orchards.

Population fluctuation was based on the total number of adults *Anastrepha* and *Ceratitis* per month, where the value obtained was determined by the sum of the number of male and female adults captured in the four weeks of the referred month, being analyzed in relation to climatic variables: temperature, relative humidity, and rainfall.

The climatic data were obtained daily by thermo hygrometers (temperature and humidity) arranged in the properties of each city, and rainfall data by the Executive Agency for the Management of Waters of the State of Paraíba - AESA.

The MAD index was calculated using the formula (captured flies number/traps installed number/collection days number) [10].

$$MAD = \frac{N}{A \times D}$$

At where:

MAD = fly/trap/day;

N = total number of captured flies;

A = number of evaluated traps;

D = interval between collections in days.

3. RESULTS AND DISCUSSION

During the study period, from July 2015 to June 2016, a total of 3,159 fruit flies were collected, of which 85.57% belonged to the genus *Anastrepha*, ~~being~~ (1.867 females and 836 males) and 14.43% ~~the species~~ *Ceratitis capitata* (Wied.), ~~being~~ (330 females and 126 males). Fruit flies were captured ~~each month on~~ all year round in eight municipalities of Paraíba swamp region, which are: Alagoa Grande (131), Alagoa Nova (471), Areia (614), Bananeiras (625), Borborema (345), Matinhas (65), Pilões (131) and Serraria (777), respectively.

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In order to evaluate the areas of domestic orchards in Paraíba swamp region, the of population fluctuation analysis was carried out using only data related to the females, where during the year a total of 10 species of the genus *Anastrepha* were captured: *A. fraterculus* (864), *A. obliqua* (535), *A. distincta* (24), *A. dissimilis* (11), *A. pickelli* (1), *A. antunesi* (37), *A. sororcula* (382), *A. zenildae* (11), *A. hadropickeli* (1), *A. barbiellinii* (1) and ~~the species~~ *C. capitata* (330).

An infestation of fruit flies was observed for both genera12 months of the year however, the genus *Anastrepha* presented ~~the~~ most of species ~~in relation~~ to the genus *Ceratitis*. The highest fruit flies population peaks occurred in April/2016 for the following species ~~and~~ municipalities: *A. obliqua* (147) in Serraria, *A. fraterculus* (91) in Areia, *A. sororcula* (58) in Bananeiras, ~~these also were species present in all municipalities every month of the year (Figure 1).~~

These population peaks occurred in the period of intense rainfall in those municipalities, where a positive correlation was observed about rainfall (r = 0.50, P> 0.05, r = 0.47, P> 0.05, r = 0.53, P> 0.05) in the tephritid populations (Table 1). During the period from December/2015 to January/2016, the lowest catch of fruit flies individuals were observed (Figure 1). This lower capture of flies is possibly due to biotic factors (natural enemies, predators, and competition) and abiotic factors (rainfall, temperature, and air humidity) which influence the life cycle of the tephritids [11].

Population peaks of fruit fly species occur according to the reproductive phenology and their host fruit maturation. According to several authors, fruit availability was a determining factor for the occurrence of fruit flies larger populations, proving that the fruiting is the phenological component that contributes the most for these insects population increase [12,13].

The population low catches are due to the unavailability of host plants for these *Tephritidae* all over the sampling period in the orchards study areas for species such as *A. distincta, A. dissimilis, A. pickelli, A. antunesi, A. zenildae, A. hadropickeli* and *A. barbiellinii*, thus justifying perhaps their low density, since some studies demonstrated that most of a certain fruit flies species remain around its preferred host [14].

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Comment [m17]: these species were also recorded monthly in all municipalities along the year (Figure 1).

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Anastrepha fraterculus was the species with the highest number of individuals collected and was present in all orchards, at all seasons of the year, having the highest catch rate in the month of September/2015. The population fluctuation graph of *A. fraterculus* was similar throughout the 12 months of the collection in practically all the municipalities, in relation to the peaks and the low population density (Figure 1). Increases in the population were observed in the fruiting and maturation periods of guava, the abundant fruit in all orchards.

About of all meteorological factors evaluated, the temperature presented the lowest correlation with fluctuation, ranging from r = -0.48 to 0.22 (*P*> 0.05). The negative correlation between temperature and fruit fly infestation in guava orchards in Mossoró, and also found that in the months where the temperature was above 28ºC there was no fruit fly infestation on fruits and trap capture rates were insignificant [10].

According to Oliveira et al. [15], climatic factors separately analyzed present little interference in the fruit fly population, indicating that these factors need to be studied together to determine their population density, a compatible fact with the results obtained in this work, where temperature and relative humidity barely interfere in the fruit flies population, but in a significant way.

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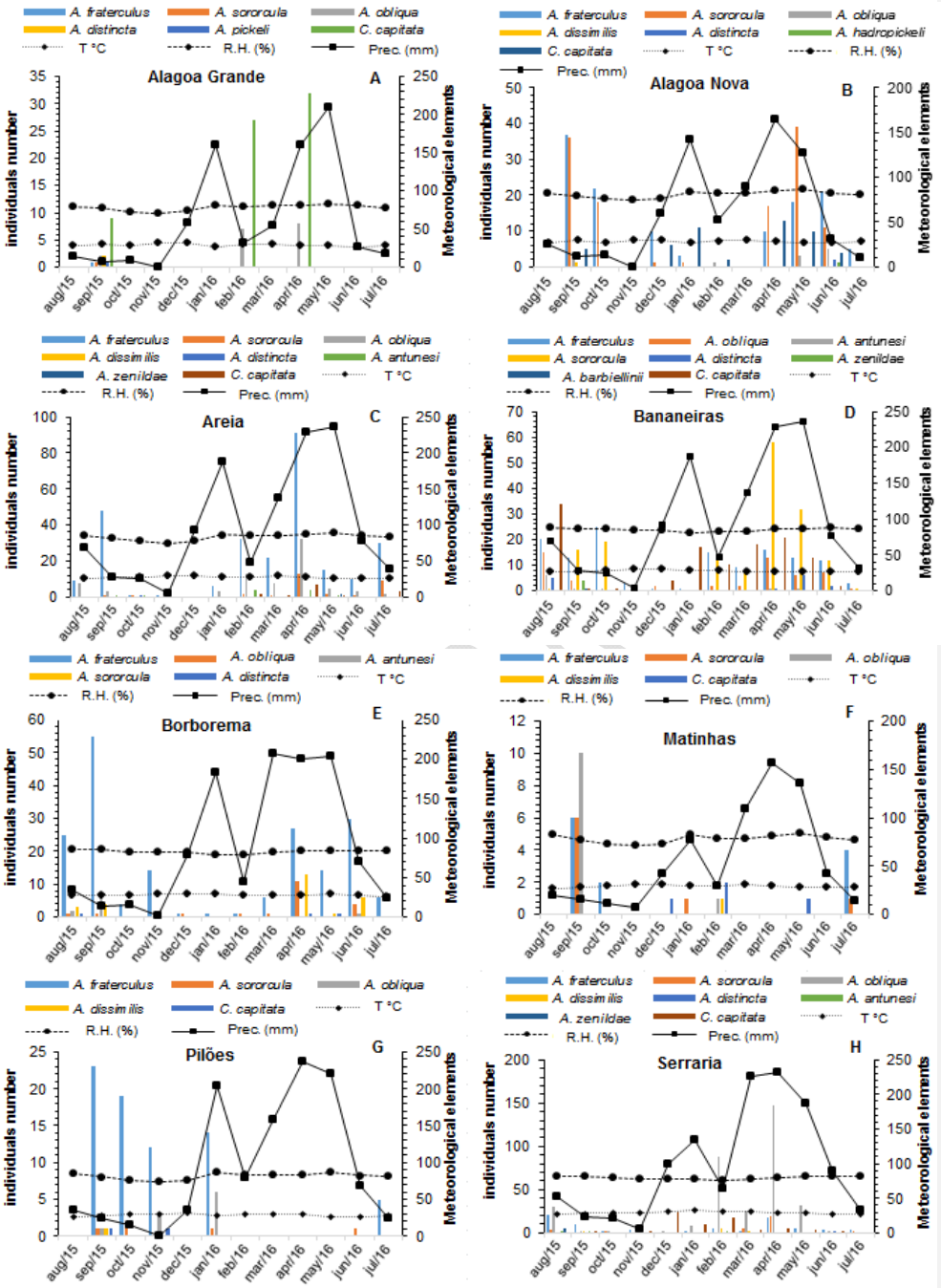

Figure 1. Fruit flies population fluctuation of *Anastrepha* and *Ceratitis capitata* species obtained from Fly-Hunting Traps in domestic orchards, from July/2015 to June/ 2016 in Paraíba swamp region

Table 1 - Pearson correlation analysis between the abiotic factors (temperature, rainfall, and relative humidity) and the average density of fruit flies trapped in orchards during the period from July/2015 to June/2016 in Paraíba swamp region

Fatores abióticos	R	T	P
	Alagoa nova		
Temperature (ºC)	-0.18 NS	-0.59	P < 0.05
Relative humidity (%)	0.18	0.59	P < 0.05
Precipitation pluvial (mm)	0.14	0.44	P < 0.05
	Alagoa Grande		
Temperature (ºC)	0.12	0.39	P < 0.05
Relative humidity (%)	0.23	0.76	P < 0.05
Precipitation pluvial (mm)	0.18	0.56	P < 0.05
	Areia		
Temperature (ºC)	-0.19 NS	-0.57	P < 0.05
Relative humidity (%)	0.46 *	1.6	P < 0.05
Precipitation pluvial (mm)	0.47 *	1.68	P < 0.05
	Bananeiras		
Temperature (ºC)	-0.48 NS	-1.73	P < 0.05
Relative humidity (%)	0.38 *	1.28	P < 0.05
Precipitation pluvial (mm)	0.53 *	1.98	P < 0.05
	Borborema		
Temperature (ºC)	-0.26 NS	-0.86	P < 0.05
Relative humidity (%)	0.66 *	2.76	P < 0.05
Precipitation pluvial (mm)	0.00	0.00	P < 0.05
	Matinhas		
Temperature (ºC)	-0.04 NS	-0.12	P < 0.05
Relative humidity (%)	-0.16 NS	-0.52	P < 0.05
Precipitation pluvial (mm)	-0.32 NS	-1.06	P < 0.05
	Pilões		
Temperature (ºC)	0.05	0.15	P < 0.05
Relative humidity (%)	-0.39 NS	-1.36	P < 0.05
Precipitation pluvial (mm)	-0.35 NS	-1.19	P < 0.05
	Serraria		
Temperature (ºC)	0.22	0.71	P < 0.05
Relative humidity (%)	-0.20 NS	-0.65	P < 0.05
Precipitation pluvial (mm)	0.50 *	1.84	P < 0.05

* Significant p <0.05.

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Was considered in the MAD index (flies/trap/day) the total number of specimens of the genera *Anastrepha* and *Ceratitis* in the orchards of eight municipalities studied.

The tephritids number collected per trap per day evidenced that during the year catches had variations in all localities and in certain samples obtained monthly the index was zero and higher than 1.0 (Figures 2). According to the specific standard for area of low prevalence of fruit flies of FAO, it defines that MAD levels established for the most diverse cultures concerning phytosanitary issues are undertaken by an NPPO (National Plant Protection Organization) of producing and importing countries, since these vary according to the susceptibility of the host and the region where the cultivation is carried out [16].

For this work, the MAD index 0.5 was used as a basis for orchards analyses, since it is the level used for decision making, indicating the need for intervention with some control method [17]. The control level in fruit orchards is established by the number of flies caught per trap per day (MAD), with the indexes being MAD 0.5, MAD 0.8 and MAD 1.0, thresholds for control through insecticides [18].

The highest rates of fruit fly catch were recorded in the months of February/2016 and April/2016. The municipality of Serraria was the city with the highest catch rate, which had an impact on the high MAD index, presenting two population peaks, one in February/2016 (MAD = 0.7) and another in April/2016 (MAD = 1.12). The above-cited fact was similar in the cities of Areia and Bananeiras, where the highest catch rates were registered in April/2016 with MAD = 1.05 and MAD = 0.6 respectively (Figure 2).

The MAD index in these localities was superior to 0.5 indicating high infestation in the orchards, with a recommendation to control fruit flies in these areas. These results are in agreement with the results found ~~by~~ [3], where MAD ranged from 0.5 to 2.0, a study conducted in the state of Amazonas. The results obtained with this study are also similar to those found ~~by~~ [19], who studied the diversity of fruit flies in the UFAM Campus, obtaining MAD from 0.89 to 3.69.

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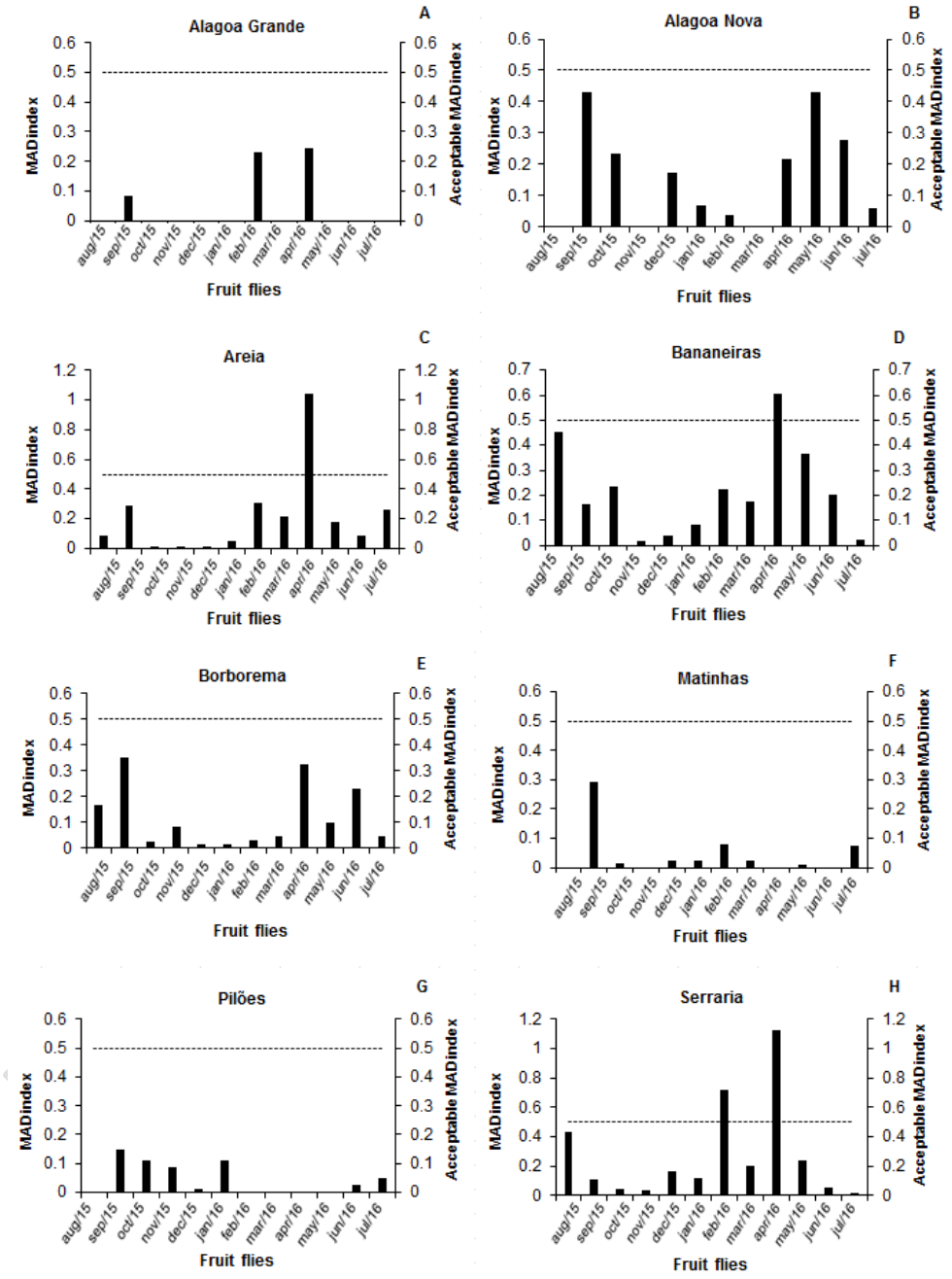
In the municipalities of Alagoa Grande, Alagoa Nova, Matinhas, Pilões and Borborema, fruit flies population fluctuation did not present a capture rate equal to or greater than 0.5 fly/trap/day at any time of the year, however the orchard located in the municipality of Alagoa Nova reached MAD levels of 0.42 and 0.43 in the months of September/2015 and May/2016 respectively, these levels found in these months are already recommended to start to carry out some measure of control for this pest. It should be noted that in this study the influence of biotic factors was no assessed, such as parasitism, predation, and competition which together with abiotic factors are responsible for the regulation of the fruit flies population fluctuation throughout the year. Fruit fly population monitoring is an important resource for integrated pest management in commercial fruit trees, allowing to verify the population fluctuation and the economic damage level [20]. The information obtained from the capture of fruit flies in traps for monitoring is important for decision-making at the beginning and at the end of control measures besides allow to evaluate the effectiveness of the strategies adopted [19].



211 **Figure 2. MAD index (flies/trap/day) of fruit flies obtained in Fly-Hunting traps in domestic**
212 **orchards, from July/2015 to June/2016, in Paraíba swamp region**
213

4. CONCLUSIONS

The low catches of certain species of fruit flies (*A. distincta, A. dissimilis*, *A. pickelli*, *A. antunesi*, *A. zenildae*, *A. hadropickeli*, *A. barbiellinii*) are due to the occurrence of alternative hosts around the orchards. The MAD index in the municipalities of Serraria, Areia and Bananeiras, in April/2016 was higher than 0.5 and it is recommended to control fruit flies in these areas. The seasonal occurrence is related to the availability of fruits, as well as the tephritid infestation rates in the domestic orchards.

REFERENCES

1. Bisognin M, Nava DE, Lisbôa H, Bisognin AZ, Garcia MS, Valgas RA, Diez-Rodríguez GI, Botton M, Antunes LEC. Biology of the South American fruit fly in blueberry, mulberry, blackberry, arachnid, and pangea fruits. Pesquisa Agropecuária Brasileira. 2013; 48 (2): 141-147. https://dx.doi.org/10.1590/S0100-204X2013000200003

2. NA Channel, Uramoto K, Zucchi RA. Two New Species of Anastrepha Schiner (Diptera Tephritidae) closely related to Anastrepha pickeli Lima. Neotropical Entomology. 2013; 42 (1): 52-57. https://doi.org/10.1007/s13744-012-0091-3

3. Ronchi-Teles B, Silva NM. Population fluctuation of species of Anastrepha Schiner. (Diptera: Tephritidae) in the region of Manaus, AM. Neotropical Entomology. 2005; 34 (5): 733-741. http://dx.doi.org/10.1590/S1519-566X2005000500004

4. Montes SNM, Raga A, Boliani AC, Santos PC. Population dynamics and incidence of fruit flies and parasitoids in peach (Prunus persica L. Batsch) cultivars in the municipality of Presidente Prudente - SP. Revista Brasileira de Fruticultura. 2011; 33 (2): 402-411. http://www.scielo.br/pdf/rbf/2011nahead/AOP04011.pdf

5. Salles LAB. Biology and life cycle of Anastrepha fraterculus. In: Malavasi A, Zucchi, RA. Fruit flies of economic importance in Brazil: basic and applied knowledge. Ribeirão Preto: Holos. 2000; 81-86.

6. Azevedo, FR, Guimarães JA, Simplício AAF, Santos HR. Faunal analysis and population fluctuation of fruit flies (Diptera: Tephritidae) in commercial orchards of guava in the region of Cariri Cearense. Archives of the Biological Institute. 2010; 77 (1): 33-41. https://www.researchgate.net/profile/Jorge_Guimaraes6/publication/266878760

7. Lemos WP, Silva RA, Araújo SCA, Oliveira ELA, Silva WR. First record of Anastrepha serpentina (Wiedemann) (Diptera: Tephritidae) in citrus in Brazil. Neotropical Entomology. 2011; 40 (6): 706-707.

8. Uramoto K. Biodiversity of fruit flies of the genus Anastrepha (Diptera: Tephritidae) at the "Luiz de Queiroz" campus, Piracicaba, São Paulo. Masters dissertation. University of São Paulo. 2002; 85.

9. Zucchi RA. Taxonomy. In: Malavasi A, Zucchi RA. Fruit flies of economic importance in Brazil: Basic and applied knowledge. Ribeirão Preto: Holos. 2000; 12-24.

10. Araújo EL, Zucchi RA. Host and levels of infestation of Neosilba pendula (Bezzi) (Diptera: Lonchaeidae) in the region of Mossoró / Assu, RN. Archives of the Biological Institute. 2002; 69 (2): 91-94. http://www.biologico.sp.gov.br/uploads/docs/arq/V69_2/araujo_II.pdf

11. Aluja M. Bionomics and management of Anastrepha. Annual Review of Entomology. 1994; 39 (1): 155-178. https://doi.org/10.1146/annurev.en.39.010194.001103

12. Alberti S, Bogus GM, Garcia FRM. Population fluctuation of fruit flies (Diptera, Tephritidae) in peach orchards and passion fruit in Iraceminha, Santa Catarina. Biotemas. 2012; 25 (2): 53-58. https://doi.org/10.5007/2175-7925.2012v25n2p53

13. Duarte RT, Galli JC, Pazini WC, Calore RA. Population fluctuation and infestation of fruit fly (Diptera: Tephritidae) as a function of the guava production system. Brazilian Journal of Agricultural Sciences. 2013; 8 (2): 241-245. https://dx.doi.org/ 10.5039 / agraria.v8i2a2717

14. Birth AS, Carvalho R, Malavasi A. Population monitoring. In: Malavasi A, Zucchi RA. Fruit flies of economic importance in Brazil: basic and applied knowledge. Ribeirão Preto: Holos. 2000; 109-112.

15. Oliveira JJD, Rocha ACP, Almeida ES, Nogueira CHF, Araújo EL. Species and population fluctuation of fruit flies in a commercial orchard of mangueira, in the coast of the state of Ceará. Caatinga Magazine. 2009; 22 (1): 222-228. https://periodicos.ufersa.edu.br/index.php/caatinga/article/view/1020

16. FAO. Establishment of areas of low prevalence for fruit. 2008. Available at: https://www.ippc.int/sites/default/files/documents/1323947574_ISPM_30_2008_En_2011-11-29_Refor.pdf. (accessed 3 November 2017).

17. Torres CAS, Castellani MA, Pérez-Maluf R, Silva JCG, Nascimento AS, São José AR, Moreira AA, Sá RF. Infestation of coffee trees by fruit flies (Diptera: Tephritidae): associated species and natural parasitism in the southwestern region of Bahia, Brazil. Applied Research & Agrotechnology. 2010; 3 (1): 135-142.

18. Rosa JMD. Determination of damage of the weevil, Conotrachelus psidii Marshall and fruit fly catch, Anastrepha fraterculus Wiedemann, with andiroba oil, Carapa guianensis in guava serrana Acca sellowiana. Masters dissertation. University of the State of Santa Catarina. 2011; 56.

19. Silva RA, Silva WR, Jesus CR, Pereira JD, Sousa Filho MF. New records of Anastrepha (Diptera: Tephritidae) for Pará. Embrapa Technical Communiqué. n.122: p, 2012.

20. Sá RF, Castellani MA, Nascimento AS, Brandão MHST, Silva AN, Pérez-Maluf R. Index of infestation and diversity of fruit flies in exotic and native hosts in the Anagé fruit stand. Bragantia. 2008; 67 (2): 401-411. http://www.scielo.br/pdf/brag/v67n2/a16v67n2.pdf