



# Phenological asynchrony between the fruit fly *Anastrepha fraterculus* and early maturing peach cultivars could contribute to pesticide use reduction

Emily S. Araujo<sup>1,3</sup>, Leticia R. Paiva<sup>2</sup>, Sidiney G. Alves<sup>2</sup>, Daniele Bevacqua<sup>3</sup>, Dori E. Nava<sup>4</sup>, Claire Lavigne<sup>3</sup> and Flávio R. M. Garcia<sup>1</sup>

<sup>1</sup>Universidade Federal de Pelotas, Dept. Ecologia, Zoologia e Genética, 96010-900, Pelotas, RS, Brazil. <sup>2</sup>Universidade Federal de São João Del-Rei, Dept. Física e Matemática, 36420-000, Ouro Branco, MG, Brazil. <sup>3</sup>INRA, UR 1115, Plantes et Systèmes de Culture Horticoles (PSH), 84000, Avignon, France. <sup>4</sup>Embrapa Clima Temperado, Laboratório de Entomologia, Rodovia BR 392, Km 78, 96010-970, Pelotas, RS, Brazil.

## Abstract

Phenological asynchrony between fruit crops and pests consists of a discrepancy between the period of fruit susceptibility and that of high pest abundance in the orchards. Therefore, it may be used for reducing pesticide applications. We assayed the potential phenological asynchrony between peach cultivars with different growing cycles and the *Anastrepha fraterculus* (Diptera: Tephritidae). To this end, we assessed fruit infestation by *A. fraterculus* at harvest for one growing season (2012-2013) in early, average and late maturing peach cultivars. Moreover, the fruit infestation was checked for non-cultivated native and non-cultivated wild exotic plant hosts around the peach orchards of the experimental area during 2013 and 2014. In addition, we monitored *A. fraterculus* abundance weekly during three consecutive growing seasons, S1 (2011-2012), S2 (2012-2013) and S3 (2013-2014), to assess phenological asynchrony between peach cultivars and *A. fraterculus*. In particular, we checked the influence of meteorological variables on *A. fraterculus* abundance, and tested if *A. fraterculus* abundance at the time when fruits are susceptible differed among cultivars. Eventually we discuss the possibility of sustainable management of peach in southern Brazil. This study constitutes a first assessment of the periods of crop vulnerability and pest presence in peach orchards in South of Brazil and provides necessary information for taking advantage of the phenological asynchrony phenomenon for this pest-crop association.

**Additional keywords:** integrated pest management; South American fruit fly; organic production; phenological resistance; *Prunus persica*; crop susceptibility.

**Authors' contributions:** DEN, FRMG, and ESA designed the study. ESA performed the experiments. CL and ESA analyzed the data. ESA, CL, DB, LRP and SGA wrote the manuscript. All authors edited, revised and approved the final manuscript.

**Citation:** Araujo, E. S.; Paiva, L. R.; Alves, S. G.; Bevacqua, D.; Nava, D. E.; Lavigne, C.; Garcia, F. R. M. (2019). Phenological asynchrony between the fruit fly *Anastrepha fraterculus* and early maturing peach cultivars could contribute to pesticide use reduction. Spanish Journal of Agricultural Research, Volume 17, Issue 1, e1001. <https://doi.org/10.5424/sjar/2019171-13294>

**Supplementary material** (Fig. S1) accompanies the paper on SJAR's website.

**Received:** 09 Apr 2018. **Accepted:** 06 Feb 2019.

**Copyright © 2019 INIA.** This is an open access article distributed under the terms of the Creative Commons Attribution 4.0 International (CC-by 4.0) License.

**Funding:** ESA received PhD scholarships from Coordination for the Improvement of Higher Education Personnel (CAPES) for the scholarships UDELAR n° 7937/13-0 and AGROPOLIS n°57/2012.

**Competing interests:** The authors have declared that no competing interests exist.

**Correspondence** should be addressed to Emily S. Araujo: [araujosemily@gmail.com](mailto:araujosemily@gmail.com)

## Introduction

Phenological asynchrony occurs when the periods of crop vulnerability and pest presence do not overlap (Messina & Jones, 1990). This condition can be induced by varying sowing time (in annual crops) or by choosing cultivars with appropriate growing cycles (in woody crops) if the pest dynamics is not too strongly dependent on that of the crop, *i.e.* the crop is not a limiting resource for the pest (Asch & Visser, 2007). Practices inducing or favoring phenological asynchrony are

important to support integrated pest management and organic farming, since they guarantee fruit production and reduce the reliance on chemical inputs and, therefore, decrease their related environmental costs (Hammons *et al.*, 2010). An assessment of the periods of crop vulnerability and pest presence is necessary to take advantage of phenological asynchrony for a given pest-crop association.

*Anastrepha fraterculus* (Wiedemann) (Diptera, Tephritidae) is a fruit pest of great economic importance because it attacks a wide variety of fruit plants and is

widely distributed, from northern Argentina to southern USA (Hernández-Ortiz & Aluja 1993). This species has a multivoltine reproduction pattern and do not have a diapause stage (Aluja & Mangan, 2008). Consequently the South America fruit fly (*A. fraterculus*) needs to infest host plants continuously to survive because this species has no mechanisms to bridge any fruit-free period (Aluja *et al.*, 2014). The females deposit their eggs in ripening fruits (Rosa *et al.*, 2017) that consequently will rot and be unmarketable (Härter *et al.*, 2015).

The Brazilian southwestern region has large extensions of natural pasture fields, with presence or absence of shrubs. In addition, there are isolated patches of native vegetation (Semi Deciduous Forest) intermixed with commercial orchards. Among the most common and widespread cultivated *A. fraterculus* hosts in the temperate region are peach (*Prunus persica* L.), apple (*Malus domestica* L.), orange (*Citrus* sp.) and grape (*Vitis* sp.), whereas native plant hosts mainly belong to the Myrtaceae Family (Nava & Botton, 2010; Machota *et al.*, 2016). The fruiting periods of all these tree species provide conditions for *A. fraterculus* to find plant hosts during all months of the year (Nava & Botton, 2010). However, abiotic factors, specifically temperature and rainfall, play an important role in tephritid outbreaks (Aluja *et al.*, 2012). Extreme thermal limits can accelerate or delay the development cycle of tephritid fruit flies (Taufer *et al.*, 2000).

The South American fly represents a threat for peach cultivars in southern Brazil (Botton *et al.*, 2002). There, the main peach-producing areas are located in the state of Rio Grande do Sul where the municipalities surrounding the city of Pelotas produce, at least, 90% of the peach national production for the canning industry (Timm *et al.*, 2007). In this region, peach orchards are commonly managed using a conventional protection strategy and trees are subjected to 4-5 broad-spectrum insecticide treatments against fruit flies per growing season. In addition, protection may be complemented by toxic baits containing hydrolyzed protein (3%) and an organophosphate insecticide (Malathion 1000 CE, 200 mL in 100 L) set on the orchard edges (Härter *et al.*, 2015).

Pest control is one of the biggest challenges for organic farming, which is increasing in Brazil but still represented less than 2% of the overall production in 2006 (IBGE, 2006). Peaches are susceptible to damage by *A. fraterculus* during their ripening period, starting ca. 30 days before harvest (Salles, 1994). In Rio Grande do Sul, grown cultivars have harvest times extending from mid-October to mid-December. Then, periods of fruit susceptibility vary depending on the cultivar and favoring phenological asynchrony with *A. fraterculus*

may be one option to reduce the application of chemical pesticides.

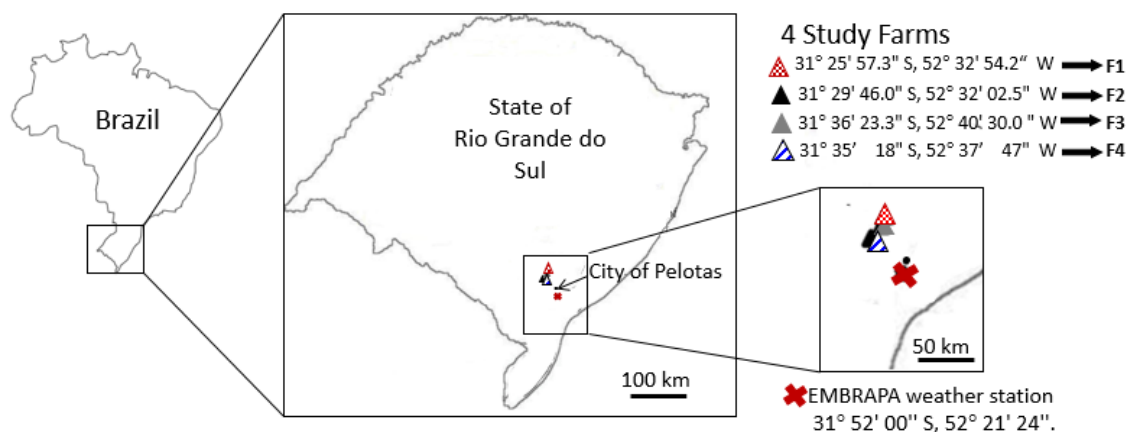
The present work aimed to assess fruit infestation by *A. fraterculus* at harvest in early, average and late maturing peach cultivars for one growing season (2012-2013). Another objective was to verify fruit infestation for non-cultivated native and non-cultivated wild exotic plant hosts around the peach orchards during 2013 and 2014. In addition, we determined the weekly abundance of *A. fraterculus* during three consecutive growing seasons, S1 (2011-2012), S2 (2012-2013) and S3 (2013-2014), in order to assess phenological asynchrony between peach cultivars and *A. fraterculus*. In particular, this study aimed to check the influence of meteorological variables on *A. fraterculus* abundance. The hypothesis to be tested was if *A. fraterculus* abundance at the time when fruits are susceptible could be different among cultivars. Eventually, we discussed the possibility of sustainable management of peach in southern Brazil.

## Material and methods

### Study area

The study was conducted in the South of the Rio Grande do Sul State, corresponding to the Pampa Biome (Fig. 1). The studied sites are located within a mosaic of seminatural vegetation, which included grasslands with shrubs and Semi Deciduous Seasonal Forest, intermixed with commercial orchards (Bilenca & Miñarro, 2004; Poppe *et al.*, 2012).

The most relevant commercial hosts grown in this region are *Prunus persica* L. (peach), *Malus domestica* Borkh. (apple), grapes (*Vitis vinifera* L. and *Vitis labrusca* L.) and *Citrus* spp. (citrus) (Bisognin *et al.*, 2015; Machota *et al.*, 2016; Rosa *et al.*, 2017). Between the most common and widespread non-cultivated fruit fly hosts in this region, plant species from the Myrtaceae family stand out, including *Eugenia uniflora* L. (Surinam cherry), *Eugenia involucrata* DC. (cereja-do-mato), *Campomanesia xanthocarpa* (Mart.) O.Berg (guabiroba), *Psidium guajava* L. (guava), *Psidium cattleianum* L. (strawberry guava); in addition, *Eriobotrya japonica* Thunb. (loquat) (Rosaceae) is quite common and abundant in the region (Salles, 1995; Bisognin *et al.*, 2015). The fruiting period of the fruit tree species present in this region provides enough availability of plant hosts for the fruit fly during all months of the year (Salles, 1995; Nava & Botton, 2010). Plant hosts of fruit flies, such as guava (fruit period from March to May), strawberry guava (fruit period from February to April) and loquat



**Figure 1.** Location and geographical coordinates (WGS84) of the four study farms (F1 to F4) and the closest EMBRAPA weather station.

(fruit period from July to September), abounded in the surroundings of the experimental area.

According to the Köppen classification system (Peel *et al.*, 2007), the study area is within the 'Cfa' (without dry season and hot summer) climate region, where average temperatures in the coldest months, July and August, are approximately 11.37 °C. During the warmest months, December and January, the average temperature is around 22.60 °C. Average daily temperature and daily rainfall were recorded from August 2011 to August 2014 at the EMBRAPA weather station closest to the studied orchards (Fig. 1).

### Fruit collection

Fruits were collected from two conventional farms of the study area, each comprising early, average and late maturing peach cultivars (Fig. 1). The first farm (F2) comprised 12.73 ha devoted to peach production, consisting of early (Precocinho), average (Granada) and late-maturing (Maciel) cultivars. In this farm, phytosanitary management was carried out using a conventional protection strategy. The second farm (F4) comprised 6 ha allocated to peach production, with early (cv. Libra), average (cv. Granada, Sensação and Esmeralda) and late (cv. Santa Aurea) maturing cultivars.

*P. persica* fruits were collected within orchards while those fruits from non-cultivated hosts were collected in non-cropped areas. First, a survey surrounding the peach orchards was carried out to identify the non-cultivated hosts present in the experimental area. This allowed identifying *Eugenia uniflora* L., *Psidium guajava* L. and *Psidium cattleianum* (Myrtaceae) and *Eriobotrya japonica* (Rosaceae) as non-cultivated plant hosts present in the experimental area. Then, fruits from these species were collected systematically during the third season

of our study (2013 to 2014) to evaluate the fruit fly infestation level according to Table 1.

Fruit infestation was calculated either as the number of pupae per fruit or as the number of pupae per kg of fruit in order to account for differences of individual fruit weight among host plants (Marsaro Júnior *et al.*, 2013). The sex ratio was calculated by dividing the number of females by the total number of individuals (males + females) (Silveira-Neto *et al.*, 1976).

Fruit samplings were adjusted to the host plant-fruiting availability. Peach sampling was carried out in 2013 at the harvest time of each cultivar. In total, 110 fruits were harvested from early-maturing cultivars on 30<sup>th</sup> October. Mid-maturing cultivars were harvested on two occasions: 198 fruits were sampled on 26<sup>th</sup> November and 1<sup>st</sup> December, at the beginning of harvest and 145 fruits were harvested on 15<sup>th</sup> and 19<sup>th</sup> December at the end of harvest. Finally, 69 fruits from late-maturing cultivars were also collected on 15<sup>th</sup> and 19<sup>th</sup> December. At each sampling date, ripen fruits were collected randomly, directly from the trees and/or the ground. The fruits were packed in bags and sent to the Insect Ecology Laboratory of the Federal University of Pelotas (Rio Grande do Sul) for species identification.

### Fruit processing, pupal recovery and fly identification

Collected fruits were placed individually in 500 mL plastic pots containing sand and vermiculite, and were covered with voile fixed with elastic tape. Each fruit remained in the pot until the larvae left them (approx. 25 days) to recover pupae, which were kept in plastic containers with vermiculite and a voile cover until the emergence of adults. The emerged insects were counted, sexed and stored individually in 70% ethanol. Each insect was taxonomically identified. The fruit

**Table 1.** Number of *Anastrepha* sp., pupae and adults, and degree of infestation, recorded for five fruit tree species surveyed in Pelotas (F2) and Morro Redondo (F4) during 2013-2014, in Rio Grande do Sul, Brazil.

Collection site	Host plant species	Sampling date	Fruits sampled	Total weight (kg) of fruits sampled	No. of pupae	No. of recovered adults	Pupae/kg of fruit	Pupae/fruit
F2	<i>Eu</i>	01/12/2013	26	0.05	0	0	0	0
F4	<i>Eu</i>	01/12/2013	51	0.36	1	0	2.77	0.02
F4	<i>Pp</i>	30/10/2013	110	82.32	0	0	0.00	0
F2; F4	<i>Pp</i>	26/11/2013	85	29	0	0	0.00	0
F2; F4	<i>Pp</i>	01/12/2013	113	10.50	0	0	0.00	0.00
F2; F4	<i>Pp</i>	15/12/2013	88	6.52	422	240	64.72	4.80
F2; F4	<i>Pp</i>	19/12/2013	126	12.01	75	35	6.24	0.60
F2	<i>Ej</i>	21/08/2014	213	1.58	476	371	301.26	2.23
F4	<i>Ej</i>	18/08/2014	465	5.75	98	98	17.04	0.21
F2	<i>Pg</i>	10/04/2014	35	2.38	211	104	88.65	6.03
F4	<i>Pc</i>	17/04/2014	41	0.23	31	31	134.78	0.75

*Eu* = *Eugenia uniflora*; *Pp* = *Prunus persica*; *Ej* = *Eriobotrya japonica*; *Pg* = *Psidium guajava*; *Pc* = *Psidium cattleianum*.

fly specimens of the genus *Anastrepha* Schiner were sexed and identified according to Steyskal (1977) and Zucchi (2000). Females were identified to the species level; males were identified only to the genus level because they do not present specific morphological characteristics (Zucchi, 2000). In the current study, only individuals from the *Anastrepha* genus were considered for subsequent analyses.

### Fruit fly monitoring

Monitoring was based upon weekly captures along three growing seasons (S1, S2 and S3), from August 2011 to August 2014, in three different farms, F1, F2 and F3 (Fig. 1) where early (Precocinho and Bonão), average (Esmeralda and Granada) and late (Eldorado, Maciel and Jubileu) maturing peach cultivars were grown and harvested on mid-October, mid-November and early December, respectively.

Twenty-seven Mcphail traps, baited with hydrolyzed protein Bioanastrepha® (5%), were installed in the orchards, three per farm and cultivar (Fig. S1 [suppl]). Details concerning the distance between traps and the orchard surface are shown in Table 2. Every 7 days, traps were cleaned, their content was renewed and the fruit

flies captured were collected and stored in 70% ethanol for taxonomic identification. Weekly trap captures were converted to the FAO phytosanitary index of flies per trap per day, calculated as  $FTD = \text{Number of captured flies} / \text{No. of traps} \times \text{Day of trap exposure}$  (IAEA, 2003) for graphical representations. For statistical analyses, fly captures per trap were summed either per month, or over the peach susceptibility period, in order to standardize the variability in trap number or capture periods (Aluja *et al.*, 2012).

### Data analysis

Linear models were used to assess if variations in monthly fly abundance depended on *i*) growing season (S1, S2 and S3), *ii*) monthly average of mean daily temperature, *iii*) monthly average of daily rainfall, and *iv*) the interaction between these two meteorological variables. Generalized mixed models with a Poisson distribution and a log link function were used to assess if the abundance of flies during the fruit susceptibility period (30 days before harvest) differed between early, average and late-maturing peach cultivars. A random 'farm' effect was included in all models to account for the nested design of the study. An individual

**Table 2.** Distance (mean  $\pm$  standard error) between traps (m) and area (ha) of the orchards in Pelotas (F1 and F2) and Morro Redondo (F3) per cultivar growing cycle.

Farm	Distance between traps (m)			Area of orchards (ha)		
	Early	Average	Late	Early	Average	Late
F1	331 $\pm$ 58.1	661 $\pm$ 124.4	143 $\pm$ 28.0	14.18	12.68	1.34
F2	236 $\pm$ 43.2	188 $\pm$ 5.3	160 $\pm$ 19.5	6.24	4.48	5.20
F3	204 $\pm$ 1.7	187 $\pm$ 36.2	174 $\pm$ 29.6	3.25	3.72	5.76

random effect was included in all models to test for over-dispersion (Harrison, 2014) and residuals were assessed visually. The significance of the fixed factors was tested by successively dropping each independent variable of the complete model (drop1 function). When a qualitative fixed factor with more than two levels was significant, pairwise multiple comparisons were performed using post-hoc Tukey tests (glht function). Statistical analyses were performed using RStudio version 1.0.143 (RStudio Team, 2016) and the packages lme4 (Bates *et al.*, 2015), Multcomp (Bretz *et al.*, 2010) and DHARMA (Hartig, 2016).

## Results

### Fruit infestation

In 2013, 77 fruits of *E. uniflora* were collected, whereas 35 fruits of *P. guajava* and 41 fruits of *P. cattleianum* were collected in 2014. In the case of the Rosaceae Family, 522 peach fruits were collected in 2013 and 678 fruits of *E. japonica* were collected in 2014 (Table 1). All flies recovered from these fruits belonged to the *Anastrepha* genus and all female flies to the *A. fraterculus* species.

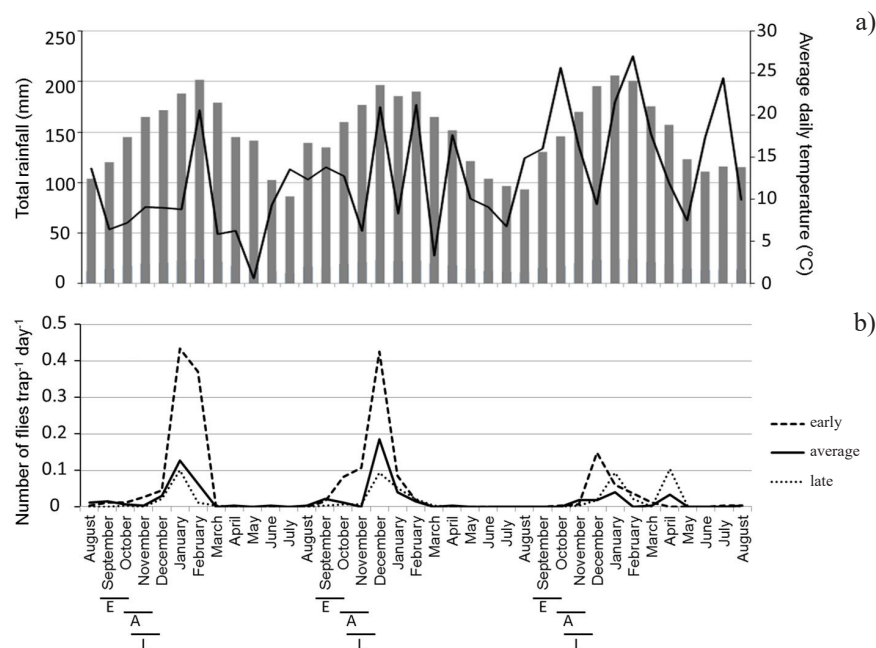
No pupae were recovered from fruits of the early or average peach cultivars collected on the first three sampling dates. In contrast, an average infestation of  $4.34 \pm 0.45$  and  $0.75 \pm 0.28$  pupae per fruit was

observed for fruits from average and late-maturing cultivars collected on the two last sampling dates. In average-maturing cultivars, 133 males and 118 females were recovered; the sex-ratio was 0.47. Concerning the late-maturing cultivars, 10 males and 14 females were recovered; the sex-ratio was 0.58.

About the native non-cultivated hosts, the highest infestation level was observed in *P. guajava* with 6.03 pupae per fruit. In this case, 104 adults were recovered, 49 males and 55 females. The sex-ratio was 0.52. During the same month, April of 2014, an infestation level of 0.75 fruit per pupae in *P. cattleianum* was verified, and 31 adults were recovered, 17 males and 14 females. The sex-ratio was 0.45. Among the exotic non-cultivated plant hosts of fruit fly, *E. japonica* had the second highest infestation level, 2.23 pupae per fruit. 678 fruits of *E. japonica* were collected, from which 469 adults were recovered, 269 males and 200 females. The sex-ratio was 0.42. A low infestation level of fruit flies was observed in *E. uniflora*, 0.02 pupae per fruit.

### Fruit fly monitoring

Over the three growing seasons, we trapped 839 Tephritidae flies, all of them belonging to the *Anastrepha* genus (61% females and 39% males). All the females were identified as *A. fraterculus*. Peaks of abundance occurred in December and January, independently of the cultivars grown in the orchards (Fig. 2). The monthly abundance of fruit flies slightly



**Figure 2.** Monthly rainfall (line, mm) and average temperature (bars, °C) in the study area between August 2011 and August 2014 (a), and fly abundance in orchards grown with early, average and late peach cultivars during the same period (b). The periods of fruit susceptibility for early (E), average (A) and late (L) maturing cultivars are indicated in the lower part of the graph.

depended on the growing season ( $F_{(3,30)} = 2.68, p = 0.06$ ), increased with the average daily temperature ( $F_{(1,30)} = 49.90, p < 0.001$ ) and decreased with average daily rainfall per month ( $F_{(1,30)} = 6.84, p = 0.014$ ). The interaction between temperature and rainfall was not significant ( $F_{(1,30)} = 0.079, p = 0.78$ ).

Total fruit fly abundance during the fruit susceptibility period differed significantly among growing seasons ( $\chi^2 = 7.4; df = 2; p = 0.02$ ), being higher in the second season than in the third one, the first season being intermediate (Table 3). It also differed significantly depending on the earliness of peach cultivars ( $\chi^2 = 14.91, df = 2; p < 0.001$ ). Fruit fly abundance was significantly higher for late-maturing peach cultivars than for those early and average-maturing (late-early:  $Z = 3.41, p = 0.002$ ; late-average:  $Z = 3.00, p = 0.007$ ), and similar for the early and average-maturing cultivars (average-early:  $Z = -0.44, p = 0.90$ ) (Fig. 3). The interaction between the effect of growing season and cultivar type on fruit fly abundance was not significant ( $\chi^2 = 3.1596; df = 4; p = 0.53$ ).

## Discussion

The current work provides a first approach for determining if phenological asynchrony could be used as a means to control pest outbursts affecting peach cultivars in South Brazil. Under these conditions, the main findings of our research were that the highest level of infestation occurred in average-maturing cultivars, while early-maturing cultivars were not infested. In addition, we detected that monthly abundance of fruit flies increased with daily temperature and decreased with monthly rainfall. Moreover, non-cultivated fruit tree species near the orchards acted as hosts for the flies. Furthermore, in the present study, *A. fraterculus* was the main fruit fly species infesting peaches in Southern Brazil in accordance with Bisognin *et al.* (2015) and Rosa *et al.* (2017). All individuals (*i.e.* females) that could be identified to the species level belonged to the *A. fraterculus* species, both in traps and among individuals that emerged from fruits. Consistently, males and females indistinctly infested fruits, as we observed sex-ratios close to 0.5.

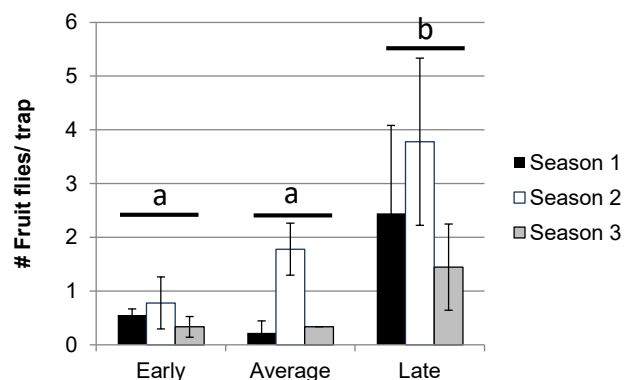
Infested fruits were recovered only from average or late maturing cultivars, harvested in mid-December. During winter (June to September), few plant hosts are available in this region. The main and most abundant plant host around the experimental area is *E. japonica*, which has the peak of fruit production in August. Our data showed a higher infestation index of 2.23 and 0.21 fruit per pupae in August of 2014. These data highlight an important

**Table 3.** Pairwise mean comparisons of *A. fraterculus* abundance in traps between the three growing seasons studied: S1 (2011-2012), S2 (2012-2013) and S3 (2013-2014) using post-hoc Tukey tests.

Growing seasons	Estimate	Standard error	z value	p (> z )
S2 - S1	0.834	0.423	1.970	0.119
S3 - S1	-0.282	0.469	-0.602	0.819
S3 - S2	-1.117	0.438	-2.546	<b>0.029</b>

Bold values indicate significant differences.

point to consider: weather conditions (temperature and relative humidity), because these are essential factors for the life cycle of insects (Hedström, 1992). The average temperature in July of 2013 and 2014 in the experimental area was 11.3 and 12.6 °C, respectively. Development and adult movements of *A. fraterculus* become faster as temperatures increase from 22 °C to 27 °C, remaining slow for temperatures below 18 °C (Salles, 1995). To complete the biological cycle (egg to adult) of the South America fruit fly takes approximately 88 days with temperatures around 15°C (Salles, 1995; Nava & Botton, 2010). If the temperature is lower, more days will be necessary to complete the biological cycle (Hedström, 1992; Nava & Botton, 2010). Therefore, if we consider that the oviposition occurred around 15<sup>th</sup> August, the adults will emerge after 15<sup>th</sup> November in the experimental area. After emergence, adults of fruit flies need to feed and they need some days to become sexually mature. This period (since egg stage until the female oviposit the first viable egg) is called period of pre-oviposition. The length of this period varies with temperature, being around 7 days at 25 - 30° C and 19 days at 20 °C. The average temperature in November of 2013 in the experimental area was 19.88 °C; this means that adults would become ready to infest fruits around the 2<sup>nd</sup> week of December, as observed in the peach sampling carried out in the experimental area.



**Figure 3.** Abundance of *Anastrepha fraterculus* in traps during the susceptibility periods of early, average and late peach cultivars during the three studied growing seasons (S1, S2 and S3). Cultivar types marked with a different letter differ significantly from each other.

The trapping results reflect the observations of peach infestation recorded in the field. In the current study, average temperatures did not reach the favorable range for *A. fraterculus* before November (Fig. 2) and, consequently, fly populations peaked in December whatever the cultivar and year. The high proportion of females obtained in the trap samples may result from the trapping methodology since protein baited traps attract more females than males due to their need to feed on protein to mature sexually and to be able to mate (Houston, 1981; Aluja *et al.*, 2012). The trapping results strongly suggest that the lower infestation in early-maturing cultivars resulted from a phenological asynchrony with fruit fly populations, since less flies were trapped during the fruit susceptibility period of early cultivars. A more favorable range of temperatures and the overlap of fruit period with different plant hosts, like *P. cattleianum* and *E. uniflora*, may have determined the higher abundance of flies late in the season. These native non-cultivated fruit trees are considered multiplying hosts of *A. fraterculus* and, thus, are sources of infestation in peach orchards (Bisognin *et al.*, 2015).

In the current study, while fruit fly populations peaked in December or January during the three studied growing seasons, their actual abundance varied from year to year, as previously observed in Southern Brazil by Rosa *et al.* (2017), and for other species of the *Anastrepha* genus in Latin America (Aluja *et al.*, 2012). In addition to direct effects of variations in climatic conditions, the abundance of fruit flies in orchards may be affected by the presence of fruiting wild host plants that may maintain or increase both fly populations and those of their natural enemies (Aluja *et al.*, 2014; Schliserman *et al.*, 2016). An interesting fact is that the peak of fruit flies captured (December-January) was particularly high in orchards with early-maturing cultivars. This should be interpreted cautiously because, although it demonstrates that *A. fraterculus* population exploited the site to obtain food and shelter, it is also possible that the traps were more attractive during that period due to the low abundance of fruits after harvest (Hendrichs & Hendrichs, 1990).

Although this study was carried out at a local scale, the results observed are relevant at a global level since early-ripening varieties of peach can suffer less attacks of tephritid flies worldwide. The fact that taking advantage of phenological asynchrony with fruit fly seasonality could be investigated in other regions and for fruit trees that have fruiting periods during the coldest months of the year, when it is too cold for tephritid fruit fly attacks. Similar results to those reported in the current study were found in peach orchards in Tunisia (Hafsi *et al.*, 2016), with a very

different climate and soil conditions than those of the current study. These authors demonstrated that early cultivars of peach suffered less attacks of *Ceratitis capitata* (Diptera: Tephritidae) than mid-maturing varieties (Hafsi *et al.*, 2016). The trapping results of this research showed that, at the beginning of the season, the number of tephritid flies were low and it increased gradually as temperatures increased to about 29 °C and as host fruits became available in nearby fields (Hafsi *et al.*, 2016). Therefore, the pattern of tephritid flies in this study was similar to that of the present survey. To demonstrate the moments when the lower densities of fruit flies occur in Southern Brazil is a great contribution to the design of integrated pest management (IPM) strategies, since some strategies for pest control, *e.g.* attract and kill bait station, are more efficient at lower densities than when the density of tephritid flies is heavy (Hafsi *et al.*, 2016). Moreover, this would be particularly important for organic peach production for which fruit flies are an important limitation in Southern Brazil (Härter *et al.*, 2015). The increase of organic crop production worldwide (Ozinci *et al.*, 2017) has raised questions about the design of specific organic crop ideotypes in breeding programs (Löschnerberger *et al.*, 2008; Crespo-Herrera & Ortiz, 2015).

Resistance to herbivores and diseases is one of the main targets for organic crops (Bruggena & Termorshuizen, 2003). Selecting for different phenologies in crops is feasible and generally aims at adapting crops to new growing regions (frost escape) (Papanikolaou *et al.*, 2005) or increasing the range of harvest dates for commercialization purposes (Raseira & Bonifacio, 2006). Few examples exist that specifically aim at phenological asynchrony with herbivores (Hammons *et al.*, 2010). In Southern Brazil, most peaches are harvested for the canning industry and late-maturing cultivars are nowadays the most suited ones for this aim because of their better texture and lesser browning susceptibility (Techakanon *et al.*, 2016). Our results indicate that a more sustainable peach farming needs that breeding programs aim at producing early-maturing peach cultivars that fulfill the aforementioned characters. As for any pest resistance that is bred into crops, evolutionary processes are expected to select pests that overcome this resistance, hereby increasing their phenological synchrony with hosts (Combes, 2001; Asch & Visser, 2007). However, since fruit flies are polyphagous and depend on a number of cultivated and wild hosts, selection for asynchrony with peach trees may not be very strong.

In summary, our results suggest that promoting phenological asynchrony between peach cultivars and

*A. fraterculus* could help to develop a more sustainable peach production and, specifically, limit insecticide use during the growing season. This study constitutes a first assessment of the periods of crop vulnerability and pest presence in peach orchards in South of Brazil and provides necessary information for taking advantage of the phenological asynchrony phenomenon for this pest-crop association. Moreover, we detected that non-cultivated fruit tree species near the orchards acted as hosts for the flies. Therefore, a deeper research on the combined effects of peach cultivar characteristics and those from other hosts on fruit fly populations will be necessary to improve their control in fruit-tree orchards.

## Acknowledgments

The authors would like to thank Prof. Lino B. Monteiro of Federal University of Paraná for engaging the collaboration between the authors and the Coordination for the Improvement of Higher Personnel (CAPES) for the scholarships. We also thank Dr. José Manuel Mirás Avalos from the University of Santiago de Compostela for his useful comments on the manuscript. We thank the useful comments from three anonymous reviewers. Also, we thank the help of Prof. Msc Maria Victoria Calvo, (UDELAR), Msc Diana Valle (INIA Las Brujas); Msc. Alexandra Peter Krüger; agronomist Ariomar A. Bartsch, and the students Marcos H. F. Telles and Victor Cieza Tarrillo for the contribution on the field and labwork. We thank in special to the agronomy student Alexandre M. Neumann for allowing us to sample peaches and other non-cultivated fruits in his farm, and by his contribution in the labwork.

## References

- Aluja M, Mangan RL, 2008. Fruit fly (Diptera: Tephritidae) host status determination: Critical conceptual, methodological, and regulatory considerations. *Annu Rev Entomol* 53: 473-502. <https://doi.org/10.1146/annurev.ento.53.103106.093350>
- Aluja M, Ordano M, Guillén L, Rull J, 2012. Understanding long-term fruit fly (Diptera: Tephritidae) population dynamics: Implications for areawide management. *J Econ Entomol* 105: 823-836. <https://doi.org/10.1603/EC11353>
- Aluja M, Sivinski J, Van Driesche R, Anzures-Dadda A, Guillén L, 2014. Pest management through tropical tree conservation. *Biodivers Conserv* 23: 831-853. <https://doi.org/10.1007/s10531-014-0636-3>
- Asch M van, Visser ME, 2007. Phenology of forest caterpillars and their host trees: the importance of synchrony. *Annu Rev Entomol* 52: 37-55. <https://doi.org/10.1146/annurev.ento.52.110405.091418>
- Bates D, Mächler M, Bolker, BM, Walker SC, 2015. Fitting linear mixed-effects models using lme4. *J Stat Softw* 67 (1), <https://doi.org/10.18637/jss.v067.i01>
- Bilenca D, Miñarro F, 2004. Identificación de áreas valiosas de pastizal (AVPs) en las Pampas y Campos de Argentina, Uruguay y sur de Brasil. Fundación Vida Silvestre Argentina, Buenos Aires, 323 pp.
- Bisognin M, Nava DE, Diez-Rodríguez GI, Valgas RA, Garcia MS, Krolow ACR, Antunes LEC, 2015. Development of *Anastrepha fraterculus* (Diptera: Tephritidae) related to the phenology of blueberry, blackberry, strawberry guava, and surinam cherry fruits. *J Econ Entomol* 108: 192-200. <https://doi.org/10.1093/jee/tou002>
- Botton M, Scoz PL, Arioli CJ, 2002. IPM on peaches in Brazil: Actual situation and future trends. *Acta Hort* 592: 655-658. <https://doi.org/10.17660/ActaHortic.2002.592.90>
- Bretz F, Hothorn T, Westfall P, 2010. Multiple comparisons using R. Chapman & Hall/CRC Press, Boca Raton, FL, USA. <https://doi.org/10.1201/9781420010909>
- Bruggena AHC van, Termorshuizen AJ, 2003. Integrated approaches to root disease management in organic farming systems. *Australas Plant Pathol* 32: 141-156. <https://doi.org/10.1071/AP03029>
- Combes C, 2001. Les associations du vivant: l'art d'être parasite. Flammarion (ed), 393 pp.
- Crespo-Herrera LA, Ortiz R, 2015. Plant breeding for organic agriculture: something new? *Agric Food Secur* 4: 25. <https://doi.org/10.1186/s40066-015-0045-1>
- Hafsi A, Abbes K, Harbi A, Duyck PF, Chermiti B, 2016. Attract-and-kill systems efficiency against *Ceratitis capitata* (Diptera: Tephritidae) and effects on non-target insects in peach orchards. *J Appl Entomol* 140: 28-36. <https://doi.org/10.1111/jen.12259>
- Hammons DL, Kurtural SK, Potter DA, 2010. Phenological resistance of grapes to the green June beetle, an obligate fruit-eating scarab. *Ann Appl Biol* 156: 271-279. <https://doi.org/10.1111/j.1744-7348.2009.00385.x>
- Harrison XA, 2014. Using observation-level random effects to model overdispersion in count data in ecology and evolution. *PeerJ* 2: e616. <https://doi.org/10.7717/peerj.616>
- Härter WR, Botton M, Nava DE, Grutzmacher AD, Gonçalves R da S, Junior RM, Bernardi D, Zanardi OZ, 2015. Toxicities and residual effects of toxic baits containing spinosad or Malathion to control the adult *Anastrepha fraterculus* (Diptera: Tephritidae). *Fla Entomol* 98: 202-208. <https://doi.org/10.1653/024.098.0135>
- Hartig F, 2016. DHARMA: residual diagnostics for hierarchical (multi-level/mixed) regression models. R package version 0.1. 0. <https://cran.r-project.org/web/packages/DHARMA/index.html> [Last access: 28 November 2018].



- Hedström I, 1992. Why do guava fruit flies, *Anastrepha striata* (Tephritidae), avoid the upper canopy of host trees? *Trop Pest Manage* 38: 136-143. <https://doi.org/10.1080/09670879209371671>
- Hendrichs J, Hendrichs MA, 1990. Mediterranean fruit fly (Diptera: Tephritidae) in nature: location and diel pattern of feeding and other activities on fruiting and nonfruiting hosts and nonhosts. *Ann Entomol Soc Am* 83: 632-641. <https://doi.org/10.1093/aesa/83.3.632>
- Hernández-Ortiz V, Aluja M, 1993. Listado de especies del género neotropical *Anastrepha* (Diptera:Tephritidae) con notas sobre su distribución y plantas hospederas. *Folia Entomologica Mexicana* 88: 89-105.
- Houston WWK, 1981. Fluctuations in numbers and the significance of the sex ratio of the Mexican fruit fly, *Anastrepha ludens* caught in Mcphail traps. *Entomol Exp Appl* 30: 140-150. <https://doi.org/10.1111/j.1570-7458.1981.tb03088.x>
- IAEA, 2003. Trapping guidelines for areawide fruit fly programmes. Insect Pest Control Section, International Atomic Energy Agency, Vienna, Austria, 48 pp. <http://www-naweb.iaea.org/nafa/ipc/public/trapping-web.pdf>
- IBGE, 2006. Censo Agropecuario. Instituto Brasileiro de Geografia e Estatística, Brasil. <https://sidra.ibge.gov.br>
- Löschenberger F, Fleck A, Grausgruber H, Hetzendorfer H, Hof G, Lafferty J, Marn M, Neumayer A, Pfaffinger G, Birschtzky J, 2008. Breeding for organic agriculture: the example of winter wheat in Austria. *Euphytica* 163: 469-480. <https://doi.org/10.1007/s10681-008-9709-2>
- Machota R, Bortoli LC, Cavalcanti FR, Botton M, Grützmacher AD, 2016. Assessment of injuries caused by *Anastrepha fraterculus* (Wied.) (Diptera: Tephritidae) on the incidence of bunch rot diseases in table grape. *Neotrop Entomol* 45: 361-368. <https://doi.org/10.1007/s13744-016-0377-y>
- Marsaro Júnior A, Deus E, Ronchi-Teles B, Adaimé R, Silva Júnior R, 2013. Species of *Anastrepha* (Diptera: Tephritidae) captured in a guava orchard (*Psidium guajava* L., Myrtaceae) in Boa Vista, Roraima, Brazil. *Braz J Biol* 73: 879-886. <https://doi.org/10.1590/S1519-69842013000400026>
- Messina FJ, Jones VP, 1990. Relationship between fruit phenology and infestation by the apple maggot in Utah. *Ann Entomol Soc Am* 83: 742-752. <https://doi.org/10.1093/aesa/83.4.742>
- Nava DE, Botton M, 2010. Bioecologia e controle de *Anastrepha fraterculus* e *Ceratitidis capitata* em pessegueiro. EMBRAPA, 29 pp.
- Ozinci Y, Perlman Y, Westrich S, 2017. Competition between organic and conventional products with different utilities and shelf lives. *Int J Prod Econ* 191: 74-84. <https://doi.org/10.1016/j.ijpe.2017.05.005>
- Papanikolaou X, Tsipouridis C, Thomidis T, Stylianidis DC, 2005. Adaptation of twenty peach and nectarine varieties in Kos and their susceptibility to Plum pox virus and *Phytophthora citrophthora*. *Hort Sci (Prague)* 32: 112-117. <https://doi.org/10.17221/3776-HORTSCI>
- Peel MC, Finlayson BL, McMahon TA, 2007. Updated world map of the Köppen-Geiger climate classification. *Meteorol Z* 11: 1633-1644.
- Poppe JL, Valente VLDS, Schmitz HJ, 2012. Structure of Drosophilidae Assemblage (Insecta, Diptera) in Pampa Biome (São Luiz Gonzaga, RS). *Pap Avulsos de Zool* 52: 185-195. <https://doi.org/10.1590/S0031-10492012001600001>
- Raseira MCB, Bonifacio H, 2006. Peach breeding program in Southern Brazil. *Acta Hort* 713: 93-97. <https://doi.org/10.17660/ActaHortic.2006.713.9>
- Rosa JM da, Arioli CJ, Dos Santos JP, Menezes-Netto AC, Botton M, 2017. Evaluation of food lures for capture and monitoring of *Anastrepha fraterculus* (Diptera: Tephritidae) on temperate fruit trees. *J Econ Entomol* 110: 995-1001. <https://doi.org/10.1093/jee/tox084>
- RStudio Team, 2016. RStudio: Integrated development for R. RStudio Inc., Boston, MA, USA. <http://www.rstudio.com/>
- Salles LAB, 1994. Periodos de ataque e de controle da mosca-das-frutas em pessegueiro. *HortiSul* 3: 47-51.
- Salles LAB, 1995. Mosca-das-frutas Sul-Americana, Empresa Brasileira de Pesquisa Agropecuária - EMBRAPA, Pelotas, Brazil. 58 pp.
- Schliserman P, Aluja M, Rull J, Ovruski SM, 2016. Temporal diversity and abundance patterns of parasitoids of fruit-infesting Tephritidae (Diptera) in the Argentinean Yungas: Implications for biological control. *Environ Entomol* 45: 1184-1198. <https://doi.org/10.1093/ee/nvw077>
- Silveira-Neto S, Nakano O, Barbin D, Villa-Nova NA, 1976. Manual de ecologia dos insetos. Piracicaba, Agronômica Ceres, 419 pp.
- Steyskal GC, 1977. Pictorial Key to Species of the Genus *Anastrepha* (Diptera: Tephritidae). Entomological Society of Washington, Washington, DC, 35pp.
- Taufer M, Nascimento JC, Cruz I, Oliveira AK, 2000. Efeito da temperatura na maturação ovariana e longevidade de *Anastrepha fraterculus* (Wied.) (Diptera: Tephritidae). *An Soc Entomol Bras* 29: 639-648. <https://doi.org/10.1590/S0301-80592000000400003>
- Techakanon C, Gradziel TM, Barrett DM, 2016. Effects of peach cultivar on enzymatic browning following cell damage from high-pressure processing. *J Agric Food Chem* 64: 7606-7614. <https://doi.org/10.1021/acs.jafc.6b01879>
- Timm LC, Reisser Junior C, Tavares VEQ, Madail JCM, Manke G, Lemos FD, Tavares LC, Radunz AL, et al., 2007. Caracterização dos persicultores irrigantes e dos métodos de irrigação no polo produtivo de pêssego da região de Pelotas. *Rev Bras Agrocienc* 13: 413-417.
- Zucchi RA, 2000. Taxonomia. pp. 13-24. In: A. Malavasi and R.A. Zucchi, (eds.). Moscas-das-frutas de importância econômica no Brasil: conhecimento básico e aplicado. Holo, Ribeirão Preto, Brazil.