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Populational dynamics of fruit flies (Diptera: Tephritidae) in guava orchards in the Northwest region of Espírito Santo, Brazil

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The diversity of fruit flies (Diptera: Tephritidae) in commercial guava orchards in the Northwest region of *Espírito Santo*, Brazil was recorded. Also, their population dynamics, correlation with climatic factors and their parasitoids were recorded. This study was carried out in three commercial guava orchards of the cultivar Paluma, located in the municipality of São Roque do Canaã, Central region of the Northwestern *Capixaba* territory, from October 2013 to September 2014. The population monitoring of fruit flies was done by installing McPhail traps provided with an attractive solution (BioAnastrepha®), hung ¾ of the guava canopy height, starting from the ground level. After obtaining the data, the following indexes were calculated: pupal viability (PV), parasitism (P) and fruits infestation (I). A correlation analysis was performed between the number of flies collected, and the meteorological variables of the region. A total of 31.51 kg of guava was collected, in which 18.31 kg were collected in the guava trees and 13.2 kg were found on the ground. 1,699 pupae were obtained, and from these, 442 flies emerged with three genera, *Anastrepha* species and one genus, *Ceratitis* (*Ceratitis capitata* Wiedemann). Only two parasitoids were obtained from the fruits from the ground, which both belong to *Doryctobracon areolatus* (Szépligeti) (*Hymenoptera: Braconidae*). The correlation analysis showed a strong correlation between the flies and fluctuating temperatures. Population peak of fruit flies occurred in February 2014.

Key words: *Psidium guajava*, Tephritidae, bioecology, parasitoids.

INTRODUCTION

The guava tree (*Psidium guajava* L.) is a fructiferous species from tropical regions and cultivated in several countries, including Brazil (Boti et al., 2016). One of the great difficulties faced by guava producers is insect attack, which infests branches, leaves and fruits, and in a short period, can cause serious problems to the crops (Gallo et al., 2002).

Insects of the Tephritidae family cause great financial losses in the fruit industries by attacking the reproductive organs of plants, fruits and flowers (Vieira et al., 2014). Fruit flies (Diptera: Tephritidae) are a serious problem every year in guava orchards, leading to increases in production cost due to frequent applications of insecticides and losses in production (Corsato, 2004).

Fruit flies present great taxonomic diversity. They comprise a complex of more than 5,000 species belonging to the Tephritidae family distributed throughout the world (Montes et al., 2011). Damage occurs due to oviposition by females in developing fruits, which causes depreciation of the product for consumption (Nunes et al., 2013). Females perforate the fruits causing cell death close to holes causing malformations in the developing fruits (Lorscheiter et al., 2012). In order to avoid this problem, chemical control is still recommended by most farmers, which is often used wrongly (Duarte et al., 2014).

One of the alternatives to reduction of the use of agrochemicals without affecting productivity is Integrated Pest Management (IPM) programs (Duarte et al., 2012). The use of McPhail traps allows verification of the population fluctuation of these insects and to relate them to the abiotic factors, especially those associated with climate, therefore helping to define which period will have a greater or lesser probability of infestations (Azevedo et al., 2010).

In the Northwest region of the state of Espírito Santo, farmers have been struggling to handle the plague in guava culture. There is lack of studies on the occurrence of this pest in guava orchards in the region. Therefore, it is of great importance to study the bioecological aspects of fruit flies in producing regions, in order to support decision-making of which management methods should be used. The objective of this study was to record the diversity of fruit flies (Diptera: Tephritidae) in commercial guava orchards in the Northwest region of Espírito Santo, Brazil, as well as its population dynamics, in correlation with climatic factors and presence of parasites.

MATERIALS AND METHODS

The field work was carried out in three commercial guava orchards of the cultivar Paluma, one hectare each, with spacing of 6 x 5 m and an average age of eight years, from October 2013 to September 2014. The areas are located in the district of *Santa Júlia*, municipality of *São Roque do Canaã*, Central region of Northwestern *Capixaba* territory (location: 19° 44 '23 "S - 40° 39' 24" W, altitude: 120 m). The average annual temperature is 23.1°C and the average annual rainfall is about 900 mm.

Population monitoring of fruit flies was done using McPhail® traps provided with 300 ml of the attractive solution based on hydrolyzed protein (BioAnastrepha®) diluted 5% and hung ¾ of the guava canopy height, starting from the ground level. The traps were hung and less exposed to the sun, and they were randomly distributed in the orchard, five per hectare (Figure 1). Renewal of the substrates from the traps and the fruit flies collection were carried out biweekly. The collected flies were identified at the genus level and stored in 70% alcohol and afterwards, the species level was indentified.

Fruit samples were collected biweekly in order to study the fruit flies species associated with guava fruits. Fruits that were present in the guava trees were collected randomly and at different canopy heights, as well as freshly fallen fruits, which were in good condition and without larvae holes (Figure 1). The samples size varied and they depended on the fruits available in the orchard.

The fruit samples were identified (date, place and person who collected them) and placed in a Styrofoam boxes and transported to the Agricultural Entomology Laboratory of the Federal University of *Santa Teresa* campus, where they were stored in plastic trays containing moist vermiculite and placed in an air-conditioned chamber at 25°C. After 10 days, the vermiculite was sieved to obtain the pupae. Afterwards, they were transferred to glass vials sealed with void tissue, containing moist vermiculite while the adults emerge. Adults were fed sucrose solution and after two to three days, the flies developed a normal color and their ovipositor matured, which was stored in 70% alcohol. When the parasitoids emerged, they were also stored in 70% alcohol for later identification.

Fruit flies and parasitoids were identified based on the keys described by Zucchi (2000) and Canal and Zucchi (2000). Genus *Anastrepha* females were collected and examined under an optical microscope (×40), according to Zucchi (2000). Data were obtained from the collected fruits on pupal viability indexes (Equation 1), parasitism (P) (Equation 2) and fruit infestation (I) (Equation 3) according to (Carvalho, 2005).

$$VP = \left[\frac{NPa + NM}{NP} \right] \text{ x } 100 \quad (1)$$

Where: PV = pupal viability; NPa = Number of emerged parasitoids; NM = Number of emerged flies; and NP = Total number of pupae obtained.

$$P = \left[\frac{NPa}{NM + NPa} \right] \text{ x } 100 \quad (2)$$

Where: P = Parasitism; NPa = number of parasitoids; and NM = number of flies.

$$I = \left[\frac{NP}{KgF} \right] \quad (3)$$

Where: I = fruits infestation; NP = number of pupae obtained; and KgF = kilogram of fruits harvested.

Data related to the adult fruit flies samples collected with McPhail® trap were plotted in frequency polygons and correlated with the meteorological data from the meteorological station of the Federal Institute of Espirito Santo - Santa Teresa Campus, where

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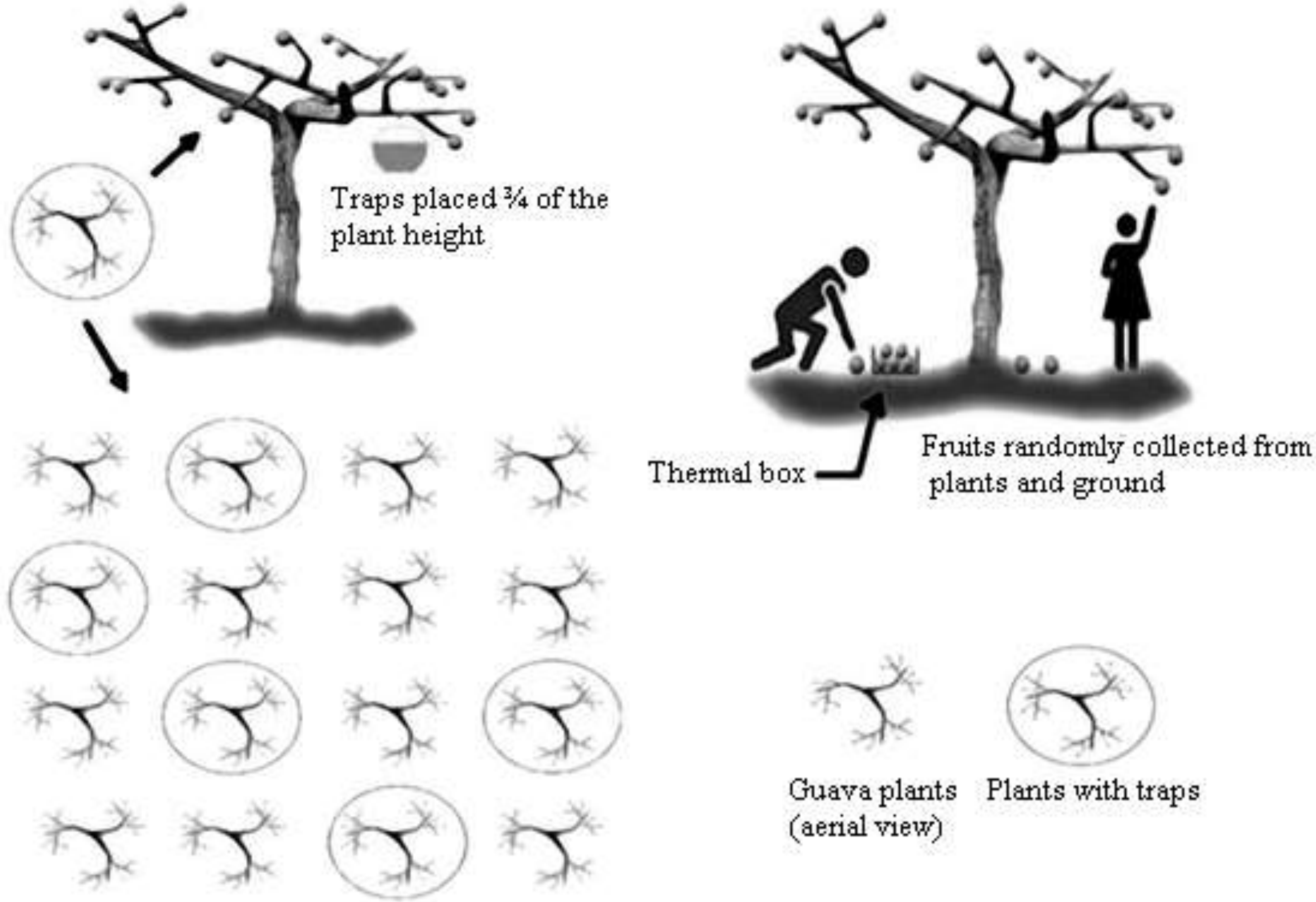

Figure 1. Scheme of traps distributed in the experimental areas (left) and the fruit sample collected (right).

temperature data, minimum temperature, average temperature, average relative humidity and rainfall are shown. Rainfall data were also obtained from rain gauges installed in the three experimental areas.

RESULTS AND DISCUSSION

In total, 31.51 kg of guava were collected of which 18.31 kg were collected from the guava trees and 13.2 kg were fruits from the ground. 1,699 pupae with pupal viability (PV) of about about 40% were obtained from the collections in the region (Table 1).

Results above these values were reported by Corsato (2004) in guava orchards in the north region of *Minas Gerais*, where they observed a pupal viability of 57.9%. Determining pupal viability is important, since the higher the value, the greater the number of individuals that could be added to the fruit fly population in the orchard.

In a study by Boff et al. (2012), in a natural guava orchard in the mountain region of *Lages* - SC, the authors found a pupal viability of 70%, a value well above the 40% observed in this study. This difference may be associated with several ecological factors, such as the orchard location, the presence of alternative hosts for fruit flies or escape areas for natural enemies, as well as the use of insecticides in orchards.

Parasitism was not observed in pupae obtained from fruits collected directly from the plants (Table 1). However, for the pupae obtained from fruits collected in the soil, parasitism was 0.43%, which corroborates with the results of Pereira-Rêgo et al. (2013), which showed that fruits collected from the ground showed greater parasitism. This is due to the infested fruits that fell on the ground having greater exposure to the parasitoids (Vargas et al., 1993). The parasitism found in this study is close to the average parasitism of 0.51% reported by Zanuncio Junior et al. (2013) in guava orchards in the municipalities of Guarapari, Serra and Viana located in the state of Espírito Santo.

From all the pupae obtained, two parasitoids were observed, both belonging to the species *Doryctobracon areolatus* (Szépligeti) (Hymenoptera: Braconidae). Considering that the main form to control tephritidae in the orchards is through the application of insecticides

Table 1. Pupal viability, parasitism and infestation index of fruit flies collected in commercial plantations in the Northwest region of *Espírito Santo*.

Collection location	*VP* (%)	*P* (%)	*I* (Puparium kg^{-1})
Ground	41.22	0.43	86.21
Plant	39.90	0.00	30.60
Total	81.12	0.43	53.92

PV: Pupal viability; P: parasitism; I: infestation index.

Table 2. Number of *Anastrepha* spp. and *Ceratitis capitata* females from the fruit collected in guava orchards in the Northwest region of Espírito Santo.

Collection location	Number of female *Anastrepha* spp. and *Ceratitis capitata*			
	A. fraterculus	*A. obliqua*	*A. zenildae*	*C. capitata*
Ground	256	4	2	10
Plant	146	15	1	8
Total	402	19	3	18
Percentage	90.95%	4.29%	0.67%	4.08%

(Härter et al., 2010), this low natural parasitism may be related to the frequent use of agrochemicals adopted as a management practice by the farmers. Araújo et al. (2015) also found a small number of parasitoids, possibly due to the drought that occurred during the study period and also insecticides that were applied in conventional orchards.

In addition to the use of agrochemicals, other factors may have contributed to the low natural parasitism found in this study, such as the host fruit and climate. Araújo et al. (2015) emphasized that the species composition of parasitoids in a region can vary considerably, depending on a series of factors such as: climate, fruit flies diversity and infested fruits, among other aspects.

The mean infestation index was 58.4 kg^{-1} puparium, being the highest infestation obtained from fruits collected from the ground (Table 1). This result reinforces the importance of crop management by removing fruits from the ground in order to reduce the fruit fly population in guava orchards.

The infestation index found in this study is higher than that found by Silva and Silva (2007), in the municipality of Ferreira Gomes - AP, where an infestation index of 5.4 kg^{-1} puparium of fruits was found. Values close to the infestation index of this study was found by Araújo and Zucchi (2003) in São Paulo, in the municipality of Santo Antônio, and the most abundant species recorded in guava orchard was *Anastrepha sororcula* (Zucchi), presenting an infestation index of 58.7 kg^{-1} puparium.

From the total adult fruit flies that emerged, the species *Anastrepha fraterculus* (Wiedemann), *Anastrepha obliqua* (Macquart), *Anastrepha zenildae* (Zucchi) and *Ceratitis capitata* (Wiedemann) were observed (Table 2). According to a survey on diversity, geographic distribution and fruit fly host conducted in the state of Espírito Santo (Martins, 2011), all species of fruit flies mentioned above already occur in the state of Espírito Santo, and in guava orchards.

The highest amount of individuals was observed for the *A. fraterculus* species (90.95%), which is also cited by Gallo et al. (1988) as the most incident in his study. Alvarenga et al. (2009) collected fruits in rural and urban areas in the municipalities of Jaíba, Janaúba and Nova Porteirinha, in northern region of the state of Minas Gerais, Brazil. They collected *C. capitata* and eight *Anastrepha* species, noting that *C. capitata* occurred mainly in introduced hosts and was prevalent in urban areas, while *Anastrepha* predominated in rural areas.

Other fruit flies species were reported in a study carried out in 2007 in the state of *Amapá* (Silva and Silva, 2007). According to the authors, the weed species were *Anastrepha striata* (Schiner), *A. fraterculus*, *A. obliqua* and *Anastrepha turpiniae* (Stone), with *A. striata* representing 76.4% of the specimens obtained. Therefore, each region of Brazil has a predominant fruit fly species, as well as infestation indexes due to different climatic conditions and available host fruits. A total of 4,475 fruit flies were collected from McPhail® traps, with the *A. fraterculus*, *Anastrepha consobrina* (Loew), *A. obliqua* and *Anastrepha grandis* (Macquart) species (Table 3), as well as the occurrence of *C. capitata*.

A. consobrina (Loew) and *A. grandis* (Macquart) species were found in the orchard with the McPhail ® trap, but they were not found in the guava fruit collected. This is because *A. grandis* (Macquart) hosts several fruits of the family, *Cucurbitaceae* (Bolzan et al., 2016) and *A. consobrina* (Loew) hosts fruits of the Passifloraceae family. Therefore, the detection of these individuals is due

Table 3. Number of female fruit flies captured with McPhail® traps in guava orchards in the Northwest region of Espirito Santo.

Number of individuals collected	Species				
	A. fraterculus	*A. obliqua*	*A. consobrina*	*A. grandis*	*C. capitata*
Total	4203	5	2	21	244
Percentage	93.94%	0.11%	0.02%	0.46%	5.45%

to the host fruits in the surroundings of the orchard.

Several *Anastrepha* species can be found in an orchard, but more than 90% are represented by one or two fly species collected in the traps (Aluja et al., 1996). This observation is confirmed in this study, where the *A. fraterculus* species represented 93.94% of the collected flies, and this is possibly due to *A. fraterculus* being one of the most polyphagous species in Brazil, with a total of 114 registered hosts (Zucchi, 2017) and can be hosted in fruits close to the orchard during the year.

The highest population densities of fruit flies occurred from December 2013 to February 2014, with population peaks in February 2014 (Figure 2), with a total of 804 flies, followed by collections in January and December, with a total of 764 and 559 flies, respectively.

The highest incidence of fruit flies occurred during fruiting season of the guava orchard, which corroborates with the results obtained by Calore et al. (2013), in a study carried out in a semi-organic orchard in the city of *Pindorama* - SP, where they verified that the greatest population peak of the flies occurred in February, and in the period of greater fruiting of the orchard.

In a study on infestation levels of *Anastrepha* spp. species in the guava crop by Araújo and Zucchi (2003), in the city of Mossoró - RN, it was verified that the highest population peaks occurred from May to July, a period that differs from the population peak found in the municipality of São Roque do Canaã - ES. However, the same authors reported that in the semi-arid regions, precipitation together with host availability is the predominant factor in population peaks, and not only the availability of fruits, in line with the results of the current study.

It is possible to observe that in April, there is a small population peak with a total of 341 flies collected, which is probably associated with host plants present in the region. This population peak occurs when Conilon coffee fruits (*Coffea canephora* Pierre ex A. Froehner) and arabic coffee (*Coffea arabica* L.) are available, and monitored orchards are located close to a plantation with two coffee species. Conilon coffee fruits available cannot be related to the population peak, because even though it is a host for tephritidae species, it presents a very low infestation index, which does not act as a natural repository for the fruit flies (Raga et al., 2002). According to Martins (2011), this low infestation is due to the fruits' physical characteristics which are, small and mesocarp has little thickness, limiting the larvae development. According to Martins (2011), this peak is possibly correlated with Arabica coffee, which has 45 times higher infestation rates than Conilon, showing that Arabica is an extremely favorable and important host as a natural repository of tephritidae.

The lowest population densities of flies occurred from May to August 2014 and during this period, few fruits were available in the orchard. This is due to several factors such as, low rainfall index and the decrease of temperature, which makes farmers to avoid pruning during this period, since the guava tends to vegetate less and produce fewer fruits. These results are comparable to those of Teles and Silva (2005), when they reported that the availability of host fruits is the most important factor in determining the occurrence and population fluctuation of fruit flies instead of the abiotic factors.

Samples were collected in all the orchards close to Atlantic forest fragments, which may have contributed to the collected flies coming from the host fruits of this biome. According to Uramoto and Martins (2005), species richness and abundance of fruit flies are higher in preserved areas than in altered ones. The period that had lowest population peaks was precisely in the months when the native vegetation of the region suffered from adverse climatic factors and consequently produced fewer fruits, which would serve as hosts for the flies.

The correlations between the meteorological factors and the population fluctuation of the fruit flies indicate that the population growth of the pest is favored at higher temperatures (Figure 2 and Table 4). On the other hand, neither rainfall nor relative humidity correlated with the number captured.

Conclusion

The species of fruit flies associated with guava fruits are *A. fraterculus, A. obliqua*, *A. zenildae* and *C.capitata*, with *A. fraterculus* being the most abundant species in the Northwest region of *Espírito Santo*.

The only parasitoid species found was *D. areolatus*; however, its parasitism index is not significant to influence the population dynamics of fruit flies.

The population peak of fruit flies in the evaluated region occurred in February. The pupal viability and the fruit flies infestation index in the studied areas were 40% and 58.4 kg^{-1} fruit puparium, respectively.

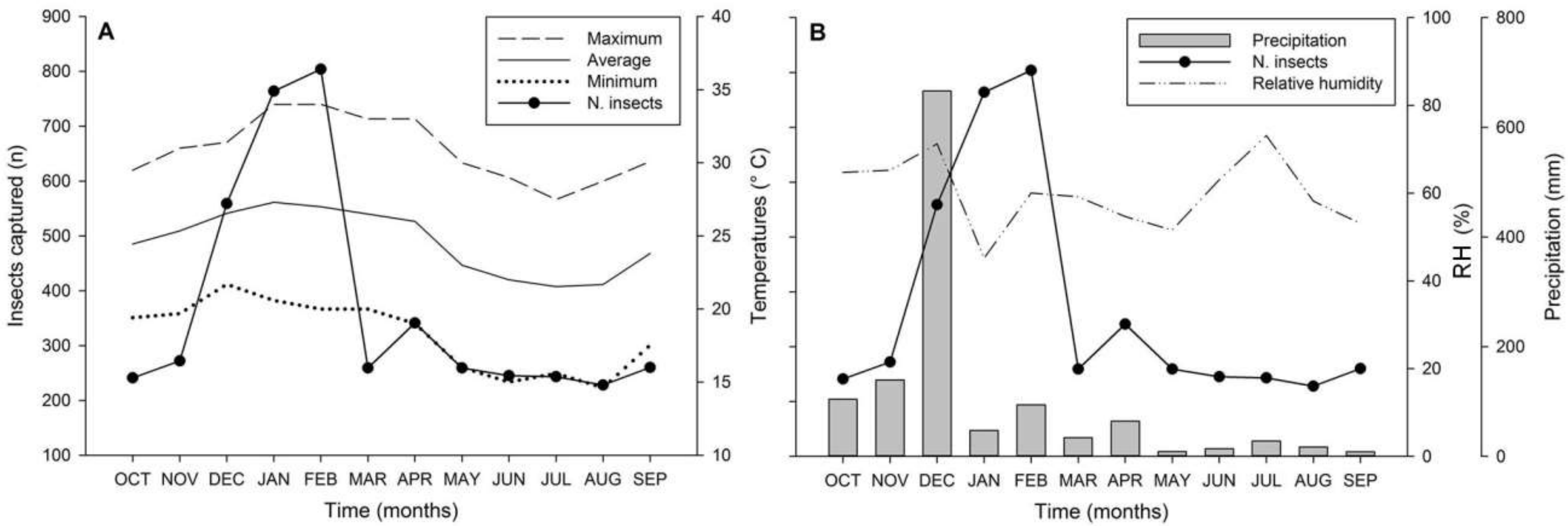

Figure 2. Population dynamics of fruit fly associated with climatic factors in guava orchards in the northwest region of Espírito Santo, from October 2013 to September 2014.

Table 4. Correlation between number of fruit flies collected in commercial guava orchards in the municipality of São Roque do Canaã - ES with McPhail® trap and meteorological factors between October 2013 and September 2014.

Meteorological factor	Number of insects captured[1]
Maximum temperature (°C)	0.733 (<0.01)
Medium temperature (°C)	0.710 (<0.01)
Minimum temperature (°C)	0.608 (<0.05)
Relative humidity (%)	-0.246 (>0.05)
Precipitation (mm)	0.327 (>0.05)

[1]Correlation of Pearson (p-value).

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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