Oviposition of Anastrepha fraterculus and Ceratitis capitata (Diptera: Tephritidae) in Citrus Fruits, and Development in Relation to Maturity of Orange Fruits

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Oviposition of *Anastrepha fraterculus* and *Ceratitis capitata* (Diptera: Tephritidae) in citrus fruits, and development in relation to maturity of orange fruits

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Abstract

Fruit flies (Diptera: Tephritidae) are considered to be one of the principal groups of pests in fruit culture worldwide. The objective of this study was to investigate oviposition by *Anastrepha fraterculus* (Wiedemann) and *Ceratitis capitata* (Wiedemann) in citrus fruits and to assess the development of these species in relation to the maturity of orange fruits. The experiments were carried out under controlled temperature conditions (25 ± 2 °C), RH (70 ± 10%), and photoperiod (12:12 h L:D). Oviposition was evaluated by exposure (with and without choice) to fruits of 'Navelina'orange [*Citrus sinensis* (L.) Osbeck], 'Clemenules' tangerine (*C. reticulata* Blanco), and 'Siciliano' lemon [*C. limon* (L.)] (Rutaceae). Insect development was studied on Navelina orange at 4 stages of maturity (I: 5 cm in diameter, II: 6 to 7 cm in diameter, III: skin with color change, and IV: yellow-orange skin). We evaluated oviposition by counting the number of eggs, and we assessed biological parameters of the immature and adult stages. The number of eggs deposited in orange and tangerine by both fruit fly species did not differ significantly, and oviposition did not occur in lemon. *Anastrepha fraterculus* preferred to oviposit in tangerine fruits, whereas *C. capitata* showed preference for orange fruits. Development from egg to adult by *A. fraterculus* and *C. capitata* occurred only in stage IV oranges, and required 30.9 and 31.0 d, respectively. Females of *A. fraterculus* and *C. capitata* developing in mature orange fruits produced a mean of 378.7 and 183.5 eggs, respectively.

Key Words: South American fruit fly; Mediterranean fruit fly; fruit fly biology; *Citrus*

Resumo

As moscas-das-frutas (Diptera: Tephritidae) são consideradas um dos principais grupos de pragas na fruticultura mundial. O objetivo deste trabalho foi verificar a oviposição de *Anastrepha fraterculus* (Wiedemann) e de *Ceratitis capitata* (Wiedemann) em frutos cítricos e avaliar o desenvolvimento destas espécies em relação ao estádio de maturação de frutos de laranjeira. Os experimentos foram realizados em condições controladas de temperatura (25 ± 2 °C), UR (70 ± 10%) e fotoperíodo (12:12 horas L:E). A oviposição foi avaliada através da exposição (com e sem chance de escolha) de frutos de laranjeira [*Citrus sinensis* (L.) Osbeck] cultivar Navelina, tangerineira (*C. reticulata* Blanco) cultivar Clemenules e limoeiro [*C. limon* (L.)] cultivar Siciliano (Rutaceae). O desenvolvimento foi estudado através da exposição de frutos de laranjeira 'Navelina' em quatro estádios (I: 5 cm de diâmetro, II: 6–7 cm de diâmetro, III: casca com mudança de coloração e IV: casca amarelo-alaranjada). Para o experimento de oviposição foi avaliado o número de ovos e para o de desenvolvimento determinou-se os parâmetros biológicos das fases imaturas e adulta. O número de ovos colocados em laranja e tangerina para ambas as espécies de mosca-das-frutas, não diferiu significativamente, sendo que em limão não ocorreu oviposição. *Anastrepha fraterculus* preferiu ovipositar em frutos de tangerineira, enquanto *C. capitata* apresentou preferência por frutos de laranjeira. O desenvolvimento de ovo-adulto de *A. fraterculus* e *C. capitata* em laranjas 'Navelina' ocorreu somente no estádio IV e teve duração de 30,90 e 31,04 dias, respectivamente. Fêmeas de *A. fraterculus* e de *C. capitata* oriundas de frutos deste estádio colocaram em média 378,7 e 183,5 ovos.

Palavras Chave: mosca-das-frutas sul-americana; mosca-do-mediterrâneo; biologia de moscas-das-frutas; *Citrus*

Fruit flies (Diptera: Tephritidae) are considered a threat to fruit orchards worldwide due to the damage caused by loss of fruit quality and to quarantine barriers imposed by importing countries (Ruiz et al. 2014). In southern Brazil, *Anastrepha fraterculus* (Wiedemann) and *Ceratitis capitata* (Wiedemann) are the 2 principal fruit fly species of agricultural importance (Nava & Botton 2010). Economic loss in citrus can reach 20% in orchards infested with *A. fraterculus* (Silva et al. 2014) and up to 7.5% in those infested with *C. capitata* (Paiva & Parra 2013).

The South American fruit fly, *A. fraterculus*, is native to South America and occurs in tropical and subtropical regions. It infests 109 plant species in Brazil (Zucchi 2008). The Mediterranean fruit fly, *C. capitata*, is indigenous to Africa and occurs worldwide. In Brazil, it infests 88 species of fruit (Zucchi 2008; Peñarrubia-María et al. 2014).

One important event in the life cycle of holometabolous phytophagous insects is choosing an oviposition site as this choice is essential for the survival and success of offspring (Segura et al. 2007). The larvae have little mobility and depend on nutritional resources selected by females at the time of oviposition (Ioannou et al. 2012). In general, insects use physical stimuli such as texture, color, shape, and size, plus chemical aspects such as nutrients, water content, attractive and repellent substances, to choose a host (López-Guillén et al. 2010).

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Tephritids have a range of hosts to choose from, and these hosts provide varying levels of physical and chemical stimuli. Monophagous species respond positively to a restricted set of volatile and visual stimuli, compared with polyphagous species (Díaz-Fleischer et al. 2000). Although this differentiation may confer an advantage to polyphagous insects, host selection does not occur without any costs because highly polyphagous females commonly oviposit in species that allow only low larval performance (Aluja & Mangan 2008). *Ceratitis capitata* provides a good example; it is considered to be one of the most polyphagous pests, as it infests approximately 300 species of fruit, including *Citrus* species (Rutaceae) (Peñarrubia-María et al. 2014).

Fruit fly infestation levels vary according to cultivar, orchard location, and climate (Ruiz et al. 2014; Silva et al. 2014). Fruit trees feature mechanisms that affect the development and reproduction of Tephritidae (Back & Pemberton 1918; Bodenheimer 1951), including changes in the physiochemical properties of fruits (Papachristos et al. 2008). However, despite the important influence of citrus hosts on the occurrence of Tephritidae, their relationship is still little studied. Because physical and chemical properties of host fruits change with maturity, this study investigated oviposition by *A. fraterculus* and *C. capitata* in orange, tangerine, and lemon fruits. We also evaluated the development of these species in orange fruits of differing maturity.

Materials and Methods

CULTURE OF *A. FRATERCULUS* AND *C. CAPITATA*

Culture and experiments were conducted in the Laboratory of Entomology of Embrapa Clima Temperado (Pelotas, Rio Grande do Sul State, Brazil), in air conditioned rooms, with temperature set at 25 ± 2 °C, RH of 70 ± 10%, and a photoperiod of 12:12 h L:D. *Anastrepha fraterculus* was reared in mango fruits (*Mangifera indica* L.; Anacardiaceae) and *C. capitata* in papaya fruits (*Carica papaya* L.; Caricaceae). Both cultures were established with wild insects obtained from fruits of tangoreiro [*Citrus sinensis* (L.) Osbeck × *C. reticulata* Blanco] cultivar 'Ortanique', collected in a commercial orchard in the municipality of Rosário do Sul (Rio Grande do Sul, Brazil) (30.1512861°S, 55.2049694°W).

Fruits were offered to the fruit flies for oviposition in wooden cages (50 × 50 × 40 cm) for a period of 24 h. Then, the fruits were packed in plastic containers (11 × 12 × 19 cm) (Sanremo, Esteio, Rio Grande do Sul, Brazil) containing a layer of finely textured vermiculite (Carolina Soil do Brasil, Santa Cruz do Sul, Rio Grande do Sul, Brazil) and topped with nonwoven fabric. At pupation, the insects were transferred to Petri dishes (10 cm in diameter × 1.5 cm high) (Kasvi, Curitiba, Paraná, Brazil) containing moistened vermiculite, where they remained until emergence. The adults were kept in wooden cages (50 × 50 × 40 cm) covered with voile fabric. Adults were provided a solid diet of sugar (União, São Paulo, Brazil), wheat germ (Walmon, São Paulo, Brazil), and yeast (Biorigin, Lençóis Paulista, São Paulo, Brazil) at the ratio of 3:1:1 in a plastic container (50 mL). In a similar container, distilled water was offered via sponge cloth (Spontex, Ilhéus, Bahia, Brazil). Water and diet were provided continuously.

FRUITS

We used fruits of 'Navelina' orange, 'Clemenules' tangerine, and 'Siciliano' lemon obtained from commercial orchards in the municipality of Rosário do Sul (30.1011111°S, 54.7172250°W and 30.1512861°S, 55.2049694°W). For the oviposition experiments, mature fruits were collected near the harvest period based on the outside color of the peel (Agustí et al. 1995). The fruits were protected from insect attack for 45 d before the collections by using wire cages (1.20 m long × 0.45 m high) wrapped with voile fabric (1.80 m long × 0.50 m high) and attached to the branches. Also, Navelina oranges were collected and used to characterize insect development in relation to fruit maturity.

OVIPOSITION IN CITRUS FRUITS: CHOICE TESTS

Females were offered 3 fruits simultaneously or combined in pairs, for a period of 24 h. Orange, tangerine, and lemon fruits were offered simultaneously using 10 cages (24 × 12 × 17 cm), each containing thirty 15-d-old females. The paired fruit combinations studied were orange and tangerine, orange and lemon, and tangerine and lemon. For paired fruit tests, we used 10 cages (24 × 12 × 17 cm) for each combination, and 20 insects (15-d-old females) were placed in each cage. After 24 h, the fruits were removed, sliced with a scalpel and inspected with a stereoscopic microscope (Zeiss, model Stemi SV 11, at 10× magnification) to count the eggs. The paired fruit experiments were conducted in a completely randomized design.

OVIPOSITION IN CITRUS FRUITS: NO-CHOICE TESTS

We provided 10 fruits of each prospective plant host in wooden cages (50 × 50 × 40 cm) to each fruit fly species for 24 h. Each cage contained one hundred 15-d-old females. The same procedure performed in the choice experiment was used to count eggs. The experiment was conducted in a completely randomized design with 10 repetitions (cages).

FRUIT MATURITY

After the orange trees flowered in 2013/14, fruit development was monitored to characterize 4 stages using methods adapted from Agustí et al. (1995). We collected 10 fruits per plant from each of 6 plants every 15 d throughout the fruiting period. The plants were identified with white ribbons fixed to the branches and the fruits were bagged in wire cages. The 4 stages were defined as: I, fruit of approximately 5.0 cm diameter (three-quarters of final size); II, fruit of 6 to 7 cm diameter (green fruit near the final size); III, fruit color changing from green to yellow; and IV, end of ripening, when the fruit is yellow-orange in color [in this stage, the ratio of total soluble solids to total titratable acidity (TSS/TTA) is ≥20].

For the physicochemical analyses, 10 fruit samples of each stage were used. In the physical analysis, we determined fruit weight (g), diameter (cm), and skin thickness (mm). The fruits were weighed on a semi-analytical scale (Shimadzu do Brasil, model BL 3200H). Fruit diameter and skin thickness were measured using a digital caliper (Stainless®, Hardened model). Chemical analyses determined the pH, total soluble solids, and total titratable acidity, and all the analyses were made in triplicate. The pH was determined using a pH meter (Phtek, model PHS 3B with Ruosull E-900 electrode). This analysis measured the pH in juice samples obtained in a microprocessor (Philips Walita, model RI7620). Total soluble solids were determined from the sample of fruit juice by using a digital Refractometer (Biobrix, model 106-D) with automatic temperature compensation, and the results were expressed in °Brix. Total titratable acidity of the juice was determined by titrations with a solution of 0.1 N NaOH and phenolphthalein indicator. Total titratable acidity results were expressed as percentage of citric acid, using the formula

$$\frac{Vg \times N\,f \times Eq.Ac.}{10 \times Va}$$

(Brasil 1986), where Vg: volume of NaOH required (mL); N: normality of NaOH solution used (0.1 N); f: correction factor obtained for standardization of NaOH = 1.00; Eq. Ac.: equivalent gram of citric acid (64.04); and Va: volume of sample (1 mL). The maturation index was calculated by the relationship Total soluble solids/Total titraable acidity.

DEVELOPMENT OF *A. FRATERCULUS* AND *C. CAPITATA*

Fruits were offered to females in wooden cages (50 × 50 × 40 cm). Six replicate cages were used for each species, each containing one hundred 15-d-old females. In each cage, 10 oranges were exposed to the flies for 24 h. Afterwards, the fruits were placed individually in plastic containers (10 × 8.5 cm) on a layer of vermiculite. The containers were closed with fine mesh. After 10 d, the fruits were checked daily, and the pupae were removed and after 24 h weighed on a precision analytical scale (Shimadzu do Brasil, model AUY 220). The pupae were then individually placed in acrylic tubes (2.5 × 4.8 × 2.5 cm) containing moist vermiculite until emergence. This procedure enabled determination of the pupal weight, duration, and viability.

After adult emergence, the sex ratio was determined and 25 individual pairs were held in cages made of 500 mL transparent plastic cups with 6 mm diameter holes on top. The pairs were fed with solid diet as described for insect culture. We also provided water in 10 mL acrylic containers.

Fecundity was determined using an artificial substrate for oviposition as described by Salles (1992). This substrate was offered to females of *A. fraterculus* and *C. capitata* and was replaced daily. We conducted daily observations to determine the number of eggs, the periods of pre-oviposition and oviposition, fecundity, and longevity of both males and females.

To assess fertility, 30 eggs from the second oviposition of each female were removed from the artificial substrates. The eggs were removed with a surgical blade and brush, and placed on a moist substrate inside Petri dishes, wrapped with PVC film, and maintained in a room at 25 °C until hatching, when the number of larvae was counted. This experiment was conducted in a randomized block design with 4 treatments (stages of maturity) and 60 repetitions (fruits). For the biology of the 2 species of fruit fly, 25 repetitions were used, each consisting of an insect pair.

STATISTICAL ANALYSES

The mean numbers of eggs in the fruit were checked for normality using the Shapiro–Wilk test (Shapiro & Wilk 1965) and outliers detected using standard deviations. The mean numbers of eggs deposited in 3-way choice tests were subjected to ANOVA and compared using Tukey's test. In 2-choice tests, the mean numbers of eggs were square root transformed ($\sqrt{x + 0.5}$) and analyzed with Student's *t* test. We also compared the influence of the presence of lemon in each combination of fruits with Student's *t* test. The physical and chemical characteristics of the oranges at 4 stages of maturity were analyzed by ANOVA and the means separated using Tukey's test.

Analyses were conducted using the statistical program BioEstat 5.3 (Ayres et al. 2007). In addition, male and female longevity data were analyzed in the statistical program JMP 5.0.1 (SAS Version 5.0.1, SAS Institute, Cary, North Carolina) using the survival analysis function. Survival curves were determined for each species of fruit fly by using the Kaplan–Meier estimator, and were compared using the log-rank test (Francis et al. 1993).

Results

OVIPOSITION IN CITRUS FRUITS

In the 3-way choice test, *A. fraterculus* preferred to oviposit in tangerine and *C. capitata* in orange. There was no oviposition in lemon fruits by either species (Table 1). This behavior was also recorded when the fruits were offered in pairs (Table 2). With respect to the potential influence of lemon on oviposition in orange or tangerine, there were no significant differences ($P = 0.4970$ and $P = 0.6633$, respectively) in the number of eggs deposited by *A. fraterculus* (Table 3). For *C. capitata*, we observed greater oviposition in orange when it was offered in combination with lemon ($P = 0.0336$) and a less significant trend (0.0975) for the same pattern with tangerine (Table 3). In no-choice tests, there was no significant difference in frequency of oviposition in orange or tangerine by either *A. fraterculus* or *C. capitata* (Table 4). In lemon, there was no oviposition by either species of fruit fly.

Table 1. Mean numbers[a] (± SD) of eggs of *Anastrepha fraterculus* and *Ceratitis capitata* recovered from citrus fruit offered simultaneously in a 3-way comparison.

Fruit	*Anastrepha fraterculus*	*Ceratitis capitata*
Orange	1.5 ± 1.5 b	3.0 ± 1.7 a
Tangerine	3.5 ± 0.9 a	1.7 ± 1.4 b
Lemon	0.0 ± 0.0 c	0.0 ± 0.0 c
Overall mean	1.7	1.5
CV (%)[b]	1.3	1.4

[a]Means within a column followed by the same lowercase letters did not differ by the Tukey test ($P > 0.05$).
[b]Coefficient of variation.

DEVELOPMENT OF *A. FRATERCULUS* AND *C. CAPITATA*

Fruit fly larvae could develop successfully only in stage IV fruits (late maturation, ratio TSS/TTA ≥ 20), when there was a significant reduction in the peel thickness, in total acidity, and consequently an increase of the ratio TSS/TTA (Table 5). Infestation indices at this stage were 0.72 and 2.05 pupae per fruit for *A. fraterculus* and *C. capitata*, respectively (Table 5).

The duration of the egg-to-adult period of *A. fraterculus* and *C. capitata* was on average 30.9 and 31.0 d, respectively (Table 6). The pre-oviposition period of *A. fraterculus* and *C. capitata* was on average 15.2 and 14.1 d, respectively, and the oviposition period lasted on average 14.2 and 18.2 d, respectively (Table 6). Most oviposition (80%) occurred in the first 17 to 20 d of the oviposition period for *A. fraterculus* and *C. capitata*, respectively, and the oviposition peak occurred on day 11 (299 eggs) for *A. fraterculus* and on day 16 for *C. capitata* (269 eggs) (Fig. 1). Females of both *A. fraterculus* and *C. capitata* survived on average for 40.5 d. Male longevity was on average 46.5 and 65.2

Table 2. Mean numbers (± SD) of eggs of *Anastrepha fraterculus* and *Ceratitis capitata* in citrus fruits offered in paired comparison tests.

Combination	Mean no. of eggs[a]	*t* value[b]	*P* value	CV (%)[c]
Anastrepha fraterculus				
Orange	1.2 ± 1.6	3.2913	0.0040	1.30
Tangerine	3.3 ± 1.6			0.49
Orange	1.8 ± 1.8	—	—	1.00
Lemon	0.0 ± 0.0			—
Tangerine	3.5 ± 1.2	—	—	0.33
Lemon	0.0 ± 0.0			—
Ceratitis capitata				
Orange	2.2 ± 1.5	2.9430	0.0087	0.70
Tangerine	0.5 ± 0.8			1.69
Orange	3.8 ± 1.1	—	—	0.29
Lemon	0.0 ± 0.0			—
Tangerine	1.7 ± 1.7	—	—	1.00
Lemon	0.0 ± 0.0			—

[a]Mean numbers of eggs were compared by the Student *t* test ($P = 0.05$).
[b]The *t* tests were not conducted where no oviposition occurred.
[c]Coefficient of variation.

Table 3. Effects of the presence of lemon on oviposition by *Anastrepha fraterculus* and *Ceratitis capitata* when offered in combination with orange or tangerine.

Combination	Mean no. of eggs[a]	*t* value	*P* value
Anastrepha fraterculus			
Orange (with lemon)	1.8 (1.8)	0.6932	0.4970
Orange (with tangerine)	1.2 (1.6)		
Tangerine (with lemon)	3.5 (1.2)	0.4426	0.6633
Tangerine (with orange)	3.3 (1.6)		
Ceratitis capitata			
Orange (with lemon)	3.8 (1.1)	2.4260	0.0336
Orange (with tangerine)	2.2 (1.5)		
Tangerine (with lemon)	1.7 (1.7)	1.7473	0.0975
Tangerine (with orange)	0.5 (0.8)		

[a]Mean numbers of eggs for each fruit, within each combination (± standard deviation), compared using the Student *t* test ($P = 0.05$).

Table 4. Mean numbers[a] (± SD) of eggs of *Anastrepha fraterculus* and *Ceratitis capitata* exposed to only 1 type of citrus fruit (no-choice tests).

Fruit	*Anastrepha fraterculus*	*Ceratitis capitata*
Orange	2.8 ± 1.9 a	5.1 ± 1.7 a
Tangerine	3.1 ± 2.0 a	5.7 ± 2.1 a
Lemon	0.0 ± 0.0 b	0.0 ± 0.0 b
Overall mean	1.9	2.8
CV (%)[b]	1.4	0.7

[a]Means within a column followed by the same letters did not differ by the Tukey test ($P > 0.05$).
[b]Coefficient of variation.

d, respectively, but no significant differences were observed between *A. fraterculus* and *C. capitata* for longevity of females ($P = 0.7382$) or males ($P = 0.6185$).

Discussion

Oviposition in choice tests for tangerine and orange indicated no influence of the presence of lemon fruit in the choice of the host by *A. fraterculus*. However, *C. capitata* females displayed a preference for oviposition in orange when lemon was present. Fruiting of Navelina orange and Siciliano lemon occurs in the same period (Feb to Jul). Therefore, there may be a preference for oviposition in orange when these citrus cultivars are found in the same site. López-Guillén et al. (2010) stated that choice for the host is influenced by visual and chemical stimuli. Thus, females of *C. capitata* may have identified compounds unsuitable for the development of their offspring in lemon. Also, Salvatore et al. (2007) found that the citrus compounds coumarin and linalool, which are found in fruits of Siciliano lemon, were toxic to larvae of *C. capitata*.

Aluja et al. (2003) reported the preference of *A. fraterculus* for tangerine fruits. However, they reported that infestation of *A. fraterculus* in orange fruits did not generate viable individuals. For *C. capitata*, Staub et al. (2008) found that 'Valencia' orange was more susceptible than 'Imperial' tangerine and Siciliano lemon, corroborating the results obtained in this study.

The preference of tephritid females for orange and tangerine fruits may be related to the suitability of the hosts for their offspring. Thus, females would prefer to oviposit in hosts that optimize larval performance (Nufio & Papaj 2004), described by Emlen (1966) as "optimal foraging."

In lemon fruits, there was no oviposition for both species of fruit fly, supporting the data obtained by Sá et al. (2008), Alvarenga et al. (2009), and Dias & Silva (2014). Although Quayle (1916) attributes the non-acceptance of lemon for oviposition by Tephritidae to fruit pulp acidity, other authors believe that lemon pulp can be suitable for larval development and the non-acceptance would be linked to volatile compounds in the fruit skin (Ruiz et al. 2014). However, Salvatore et al. (2007) artificially infested Siciliano lemon with eggs of *C. capitata* and obtained up to 98% mortality of larvae. The authors attributed the high mortality to the combined action of egg encapsulation in essential oil glands of the flavedo and the toxicity of the albedo. However, the relationship of fruit acidity with the development of fruit flies has not yet been demonstrated.

Our data on infestation of mature Navelina orange fruits confirm the results obtained by Greany et al. (1985), showing that unripe fruits do not provide ideal conditions for development of tephritid larvae. Silva et al. (2014) and Dias et al. (2013) reported similar results with *A. fraterculus* in orange (0.86 pupae per fruit). It was evident that there was greater susceptibility of mature orange, especially to *C. capitata*, which supports the work of Joaquim-Bravo et al. (2001), Lopes et al. (2009), and Arredondo et al. (2015).

Overall, both orange and tangerine fruits were accepted for oviposition by *A. fraterculus* and *C. capitata*, but lemon fruits were not. *Anastrepha fraterculus* showed preference for oviposition in Clemenules

Table 5. Mean values (± SD) of physical and chemical characteristics of orange, and the infestation index (IF) of *Anastrepha fraterculus* (A.f.) and of *Ceratitis capitata* (C.c.) in 4 maturation stages. Agricultural crop 2013/14.

								IF	
Stage[a]	Weight	Diameter	Thickness	pH of juice	TSS[b]	TTA[c]	Ratio TSS/TTA	A.f.	C.c.
I	55.03 ± 5.11 c (49.43–67.00)	4.74 ± 0.19 c (4.46–5.01)	7.68 ± 0.71 a (6.63–8.71)	2.85 ± 0.09 c (2.71–3.04)	9.90 ± 0.88 c (8.60–11.30)	0.84 ± 0.01 a (0.83–0.86)	11.75 ± 1.07 d (9.95–13.27)	—	—
II	120.04 ± 8.02 b (106.78–132.39)	6.19 ± 0.24 b (5.86–6.67)	4.43 ± 0.28 b (4.03–4.99)	3.12 ± 0.09 b (2.99–3.23)	10.20 ± 0.30 bc (9.90–10.90)	0.78 ± 0.01 b (0.76–0.79)	13.04 ± 0.34 c (12.47–13.73)	—	—
III	245.87 ± 22.45 a (210.80–284.07)	7.80 ± 0.27 a (7.35–8.30)	3.48 ± 0.34 c (3.03–3.99)	3.56 ± 0.14 a (3.32–3.73)	10.7 ± 0.50 ab (10.00–11.40)	0.59 ± 0.00 c (0.59–0.61)	17.95 ± 0.99 b (16.44–19.18)	—	—
IV	257.30 ± 23.89 a (217.37–281.89)	7.98 ± 0.32 a (7.36–8.43)	2.49 ± 0.37 d (1.82–2.97)	3.56 ± 0.05 a (3.48–3.66)	11.10 ± 0.25 a (10.80–11.60)	0.52 ± 0.00 d (0.51–0.54)	21.27 ± 0.70 a (20.08–22.17)	0.72	2.05
CV (%)[d]	10.07	3.97	10.20	3.10	5.19	1.50	5.18	—	—

Values in a column followed by the same letter did not differ significantly using Tukey's test ($P > 0.05$). Values in parentheses indicate the range.
[a]Stages: I, fruit of approximately 5.0 cm diameter; II, fruit of 6.0 to 7.0 cm diameter; III, green fruit changing to yellow; and IV, mature orange fruit (TSS/TTA ≥ 20).
[b]Total soluble solids.
[c]Total titratable acidity, expressed as a percentage of citric acid.
[d]Coefficient of variation.

Table 6. Mean values (± SD) and range of biological parameters of immatures and adults of *Anastrepha fraterculus* and *Ceratitis capitata* when developing in stage IV oranges.

Biological parameter	*Anastrepha fraterculus*	*Ceratitis capitata*	CV (%)[a]
Period egg to adult (d)	30.9 ± 1.30 (29.0–32.0)	31.0 ± 2.01 (28.0–37.0)	5.34
Weight of pupae (mg)	12.6 ± 0.31 (11.8–12.9)	11.5 ± 0.53 (10.2–13.0)	3.55
Number of pupae	0.7 ± 1.35 (1.0–5.0)	2.1 ± 3.18 (1.0–13.0)	17.21
Viability of pupae (%)	61.1 ± 30.46 (25.0–100.0)	70.3 ± 19.60 (50.0–100.0)	38.88
Sex ratio (female/male)	0.48	0.50	—
Pre-oviposition period (d)	15.2 ± 1.54 (13.0–18.0)	14.1 ± 2.64 (8.0–21.0)	14.45
Oviposition period (d)	14.2 ± 2.94 (10.0–19.0)	18.2 ± 4.60 (10.0–25.0)	22.86
Fecundity (number of eggs)	378.7 ± 61.54 (306.0–473.0)	183.5 ± 56.08 (106.0–294.0)	23.40
Fertility (%)	60.4 ± 7.48 (43.8–69.2)	62.0 ± 5.20 (50.0–67.7)	10.38

Values in parentheses indicate the range.
[a]Coefficient of variation.

tangerine fruits, whereas *C. capitata* preferred Navelina orange fruits. Based on the infestation of fruit flies determined in orange, both species preferred to oviposit in ripe fruits.

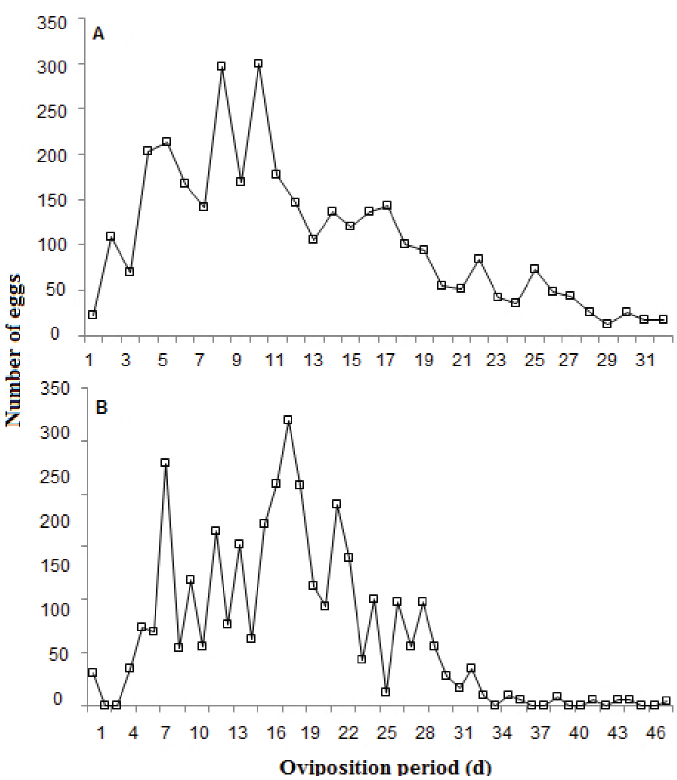

Fig. 1. Frequency of oviposition by females of *Anastrepha fraterculus* (A) and *Ceratitis capitata* (B) when provided with oranges as an oviposition substrate.

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