BIOLOGICAL CONTROL

Diapause in Fruit Fly Parasitoids in the Recôncavo Baiano, Brazil

Romulo da S. Carvalho

Embrapa Mandioca e Fruticultura, C. postal 007, 44.380-000, Cruz das Almas, BA, romulo@cnpmf.embrapa.br

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Diapausa em Parasitóides de Moscas-das-Frutas no Recôncavo Baiano

RESUMO - A diapausa tem sido pouco estudada em moscas-das-frutas e seus parasitóides em regiões de clima tropical. Este trabalho é o primeiro registro da diapausa em parasitóides de larva-pupa de tefritídeos no Recôncavo Bajano. A diapausa foi observada em quatro espécies nativas [Dorvctobracon areolatus (Szépligeti), Utetes anastrephae (Viereck), Opius sp. (Hymenoptera: Braconidae), Aganaspis pelleranoi (Brethes) (Hymenoptera: Figitidae)] e uma introduzida [Diachasmimorpha longicaudata Ashmead (Hymenoptera: Braconidae)]. Os parasitóides diapáusicos foram obtidos de pupários provenientes de frutos coletados de novembro/1998 a abril/2000. Sua freqüência total foi de 1,4%, sendo D. areolatus a espécie mais freqüente, com tempo de desenvolvimento variando de 82 a 414 dias após a coleta dos frutos. Nos pupários provenientes de goiaba e serigüela, somente D. areolatus e A. pelleranoi foram observadas em diapausa. Em goiaba, a emergência dos adultos de A. pelleranoi foi de 222 e 263 dias e a de D. areolatus 82 e 170 dias. Em serigüela, D. areolatus emergiu entre 157 e 327 dias. Pitanga foi a fruteira com o maior número de espécies de parasitóides diapáusicos. Nessa fruteira, D. areolatus foi a mais freqüente com emergência entre 82 e 414 dias. Foram observadas também U. anastrephae (277 dias), Opius sp. (243 dias), D. longicaudata (294 dias) e A. pelleranoi (270 e 305 dias). Em carambola, D. areolatus emergiu entre 150 e 190 dias após a coleta dos frutos e D. longicaudata 164 e 216 dias. Em jambo, D. areolatus emergiu com 181 e 314 dias e 269 dias para a manga.

PALAVRAS-CHAVE: Controle biológico, Tephritidae, Braconidae, Figitidae, dormência

ABSTRACT - Diapause has been poorly investigated in tephritid fruit flies and in associated parasitoids in tropical regions. In this work, diapause in larval-pupal parasitoids of fruit flies is reported for the region of the Reconcavo Baiano, Brazil. Diapause was recorded for individuals of four native species [Doryctobracon areolatus (Szépligeti), Utetes anastrephae (Viereck), Opius sp. (Hymenoptera: Braconidae), Aganaspis pelleranoi (Brethes) (Hymenoptera: Figitidae)] and of one exotic braconid [Diachasmimorpha longicaudata Ashmead]. Diapausing parasitoids were obtained from fruit fly puparia in fruits collected between November 1998 and April 2000. In all, 1.4% of individuals underwent diapause, being D. areolatus the most frequent. The development time for this species ranged from 82 to 414 days. In puparia obtained from guava and hog plum, only D. areolatus and A. pelleranoi went through diapause. In guava, total development time for A. pelleranoi varied from 222 to 263 days and for D. areolatus from 82 to 170 days. In hog plum, D. areolatus emerged after a period of 157 to 327 days. Brazilian cherry was the species with highest number of diapausing parasitoid species, being D. areolatus the most abundant and emerging at 82-414 days after fruit collection. U. anastrephae (277 days), Opius sp. (243 days), D. longicaudata (294 days) and A. pelleranoi (270 and 305 days) were also observed. In carambola, D. areolatus adults emerged 150 and 190 days after fruit collection and *D. longicaudata* between 164 and 216 days. In water apple *D. areolatus* emerged between 181 and 314 days and 269 days in mango.

KEY WORDS: Biological control, Tephritidae, Braconidae, Figitidae, dormancy

Insects are highly adaptable to environmental conditions. Synchronization of their activities to favorable periods and increase of the likelihood of survivorship during unfavorable ones is achieved by individuals of several species through a dormancy period. The term "dormancy" includes quiescence as well as diapause (Delinger 1986). Shelford (1929) introduced the term quiescence to describe the delay in development related to environmental conditions. The term diapause was proposed by Wheeler (1893) to describe a specific embryonary stage during which the embryo interrupts its development before the final movements of blastokinesis. Diapause involves a much more pronounced paralisation in insect development.

Diapause is generally considered and adaptive strategy developed by insects that inhabit regions of temperate climate, as development is not possible during winter. According to Denlinger (1986), diapause may also occur in insects that inhabit tropical environments, though factors that regulate the phenomenon have not been fully understood yet. In tropical regions, diapause enables insects to survive during dry season, characterized by the lack of food.

Diapause in fruit fly parasitoids has been reported in temperate regions (Prokopy 1968, Maier 1981, AliNiazee 1985, Hoffmeister 1992, Gut & Brunner 1994). In tropical environments, this phenomenon has been poorly studied in fruit fly and associated parasitoids (Aluja et al. 1998). The first evidence for diapause in larval-pupal parasitoids of fruit flies in tropical environments was provided by Pemberton & Willard (1918). Authors referred to "hibernation' in Biosteres (Diachasma) tryoni (Cameron) and Opius fullawaui (Silvestri). Later, Darby & Knapp (1934) found diapausing individuals of Doryctobracon (= Opius, Diachasma) crawfordi (Viereck) infesting Anastrepha ludens (Loew). These authors observed that some individuals emerged up to seven months after pupation, and presumed that this was due to diapause.

Clausen *et al.* (1965) observed diapause in Diachasmimorpha (= Biosteres) longicaudata, Opius formosanus Fullaway, O. compensans Silvestri (both synonyms of D. longicaudata); O. watersi Cameron (= Diachasmimorpha dacusii). Ashley *et al.* (1976) observed that diapause in D. longicaudata is affected by low temperatures and humidity in pupation substrate. Aluja *et al.* (1998) obtained ecological evidence for diapause in two native species of Braconidae [Doryctobracon areolatus (Szépligeti) and Utetes (B.) anastrephae (Viereck)], as well as in an exotic species (D. longicaudata) and two eucoilines (Figitidae), namely Aganaspis pelleranoi (Brethes) and Odontosema anastrephae (Borgmeier).

The present work reports the occurrence of diapause in four native species of fruit fly parasitoids and in the exotic parasitoid *D. longicaudata* in the region of Recôncavo Baiano.

Material and Methods

Collection Area. Fruits were collected in Conceição do Almeida, BA, in the region of the Recôncavo Baiano (12°48'45"S, 39°15'20"W, 190 m altitude) in the Estação de Fruticultura Tropical of the Empresa Baiana de Desenvolvimento Agropecuário – EBDA. Fruits of the following species were collected underneath the canopy of trees from November 1998 to April 2000: Brazilian cherry -*Eugenia uniflora* L. (Myrtaceae); carambola - *Averrhoa carambola* L. (Oxalidaceae); guava - *Psidium guajava* L. (Myrtaceae), hog plum - *Spondias purpurea* L. (Anacardiaceae), mango - *Mangifera indica* L. (Anacardiaceae); water apple - *Syzygium aqueum* Burm. F. (Alston) (Myrtaceae) and malay rose apple - *Syzygium malaccense* L. (Myrtaceae).

Collection of Fruit Fly Puparia and Emergence Data. In the laboratory, fruits were counted and placed in plastic boxes containing vermiculite as pupation medium. Fruit fly puparia were sieved, counted and placed in glass vials containing vermiculite and closed with a thin-mesh tissue. Following adult emergence that usually occurred 10-15 days after pupation, uneclosed puparia were separated and maintained at laboratory conditions ($26 \pm 2^{\circ}$ C, RH 70 \pm 10% and photophase 12h). Daily observations on adult emergence were undertaken.

Evaluation of Pupal Mortality Factors. The total of 752 uneclosed puparia were randomly taken from samples, which were dissected under the steromicroscope. The following mortality factors were considered: a) disease: number of puparia infected by fungi (pupae containing micelia in their interior or external surface) and bacteria (dark pupae with disintegrated tissue and characteristic odor); b) dehydration: number of puparia containing dry insects inside and c) empty puparia: number of swallow puparia.

Parasitoid Identification. Braconids were identified according to Wharton & Giltrap (1983) and Canal Daza (1994). *Aganaspis pelleranoi* (Brèthes) Eucoilinae (Figitidae) was identified in the Setor de Entomologia of ESALQ/USP.

Results and Discussion

Out of the 12,926 fruits collected, 23,349 puparia were obtained, from which 11,104 fruit flies emerged being 97.8% representatives of the genus *Anastrepha* and 2.2% of *Ceratitis capitata* (Wied.). The total of 2,538 parasitoids emerged at the normal range of development and 191 (1.4%) were diapausing individuals.

The native braconid *D. areolatus* represented 92.2% of the total number of diapausing parasitoids (Table 1). Diapause frequency in *D. areolatus* was 65.5% in Brazilian cherry, 16.8% in hog plum, 3.7% in carambola, 3.1% in guava, 1.6% in malay rose apple and 1.0% in water apple.

According to Bateman (1972), in tropical environments, at the beginning of the fruiting period, the abundance of fruit flies is low. Usually, fruit fly populations build up quickly and yield large numbers of descendents. When host supply decreases, so does the fruit fly population. According to Dellinger (1986), some fruit fly species synchronize their emergence period with the period of host fruit availability, as a survival strategy. In this manner, parasitoids also

Parasitoid species/ host plant	Number of diapausing individuals	Diapause duration (days)		% of
		Min.	Max.	diapausing individuals
D. areolatus				
Carambola - A. carambola	7	150	216	3.7
Guava - P. guajava	6	82	263	3.1
Water apple - S. aqueum	2	181	302	1.0
Malay rose apple - S. malaccense	3	293	343	1.6
Mango - M. indica	1	-	269	0.5
Brazilian cherry - E. uniflora	125	111	414	65.5
Hog plum - S. purpurea	32	157	327	16.8
D. longicaudata				
Water apple - S. aqueum	1	-	181	0.5
Carambola - A. carambola	2	164	216	1.1
Brazilian cherry - E. uniflora	1	-	294	0.5
A. pelleranoi				
Guava - P. guajava	6	222	263	3.1
Brazilian cherry - E. uniflora	2	270	305	1.1
Hog plum - S. cirouella	1	-	298	0.5
Opius sp.				
Brazilian cherry - E. uniflora	1	-	243	0.5
U. anastrephae				
Brazilian cherry - E. uniflora	1	-	277	0.5

Table1. Frequency, duration of the diapausing period and number of native and introduced fruit fly parasitoids that diapaused in different host plants. Conceição do Almeida, BA, 1998-2000.

synchronize their emergence with host availability. In the present work, even though emergence of diapausing parasitoids occurred all along the year, most of them emerged during the period of higher host fruit availability, i.e. from September to March (Fig. 1).

Aluja et al. (1998) reported that in Mexico, diapausing individuals of the following fruit fly parasitoid species were observed from September to December in localities higher than 1,000 m above sea level: D. areolatus (native), U. anastrephae (native), A. pelleranoi (native), Odontosema anastrephae Borgmeier 1935 (Eucoilinae) (native) and D. longicaudata (exotic). According to these authors, D. areolatus diapause period was up to 10 months, being recovered from fruit fly larvae obtained in Spondias mombin L. A. pelleranoi was recovered from P. guajava and diapause was as long as 11 months. The same period was observed for U. anastrephae reared from S. mombin. Also, the shortest periods were reported for D. longicaudata in S. mombin (4 months), P. guajava (3 to 6 months), Psidium sartorianum (O. Berg) Nied. (3 months) e Citrus sinensis (L.) Osbeck (3 months).

The frequency of diapausing parasitoids observed in all fruit species sampled in the present study was 1.4%. Longest diapause duration was observed in *D. areolatus* (3 to 14 months), followed by A. *pelleranoi* (7-10 months), *D. longicaudata* (5-10 months), *U. anastrephae* (9 months) and *Opius* sp. (8 months) (Table 1). Our data show that diapause duration is highly variable, but thus far, causes that lead to

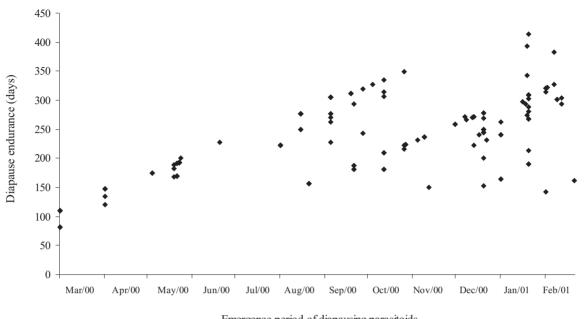
this heterogeneity have not been elucidated.

The diapausing individuals of *D. areolatus* were obtained from fruits of water apple e malay rose apple, carambola, guava, mango, Brazilian cherry and hog plum. Our data corroborate the conclusions of Aluja *et al.* (1998), as *D. areolatus* was the species with longest diapause period (414 days).

Diapausing individuals of *A. pelleranoi* were obtained from guava, Brazilian cherry and hog plum, being guava the host with highest frequency of diapause (3.1%). The longest diapause period was observed in parasitoids obtained from Brazilian cherry (305 days), followed by hog plum (298 days) and guava (263 days).

The diapausing individuals of the native braconids *Opius* sp. and *U. anastrephae* were obtained exclusively from Brazilian cherry. Diapause duration ranged from 243 dias to 277 days, respectively. Diapausing individuals of the exotic *D. longicaudata* were obtained from water apple, carambola and pitanga, with maximum duration of 181, 216 and 294 days. Though represented in the collection area, no diapausing individuals of *Asobara anastrephae* (Muesebeck) (Alysiinae) were obtained.

D. areolatus, regardless of host fruit, showed the highest number of diapausing species, representing 92.2% of all diapausing individuals collected (Table 1). The minimum duration of diapause for this species was 110 days and the maximum 414 days for individuals obtained from Brazilian cherry. In hog plum, this period varied from 157 to 327 days. Brazilian cherry stands out also as the host species with



Emergence period of diapausing parasitoids

Figure 1. Emergence period of the native and exotic parasitoids under laboratory conditions. Cruz das Almas, BA, Brazil, 2000-2001.

highest number of diapausing parasitoids and frequency (65.5%).

The eucoiliine *A. pelleranoi* was the second species in terms of percentage of diapausing individuals (total frequency of 4.7%), being 3.1% in puparia obtained from guavas, 1.1% from Brazilian cherry and 0.5% from hog plum. Diapause duration for *A. pelleranoi* ranged from 222 to 305 days, being Brazilian cherry the host with longest diapause period (Tabela 1).

The braconid *D. longicaudata* was the third in frequency of diapausing individuals: 2.1%. Individuals obtained from carambola showed the highest frequency of diapause (1.1%), followed by water apple and Brazilian cherry, both with 0.5%. The duration of diapause for *D. longicaudata* varied from 164 to 294 days. The Brazilian cherry was the host where the longest diapause period was registered for this species (294 dias).

Brazilian cherry was the only host plant where diapausing individuals of *Opius* sp. and *U. anastrephae* were recovered. *Opius* sp. and *U. anastrephae* showed the same frequency of diapausing individuals (0.5%) and longest diapause duration was 243 and 277 days, respectively (Table 1).

From 42.4% of the puparia obtained in different host species no fruit flies or parasitoids emerged. Possible mortality factors were dehydration (63.7%), fungi (13.8%), bacteria (0.4%) and diapause (0.5%) (Fig. 2). Also, 21.5% of the puparia were empty due to unidentified causes.

Bressan-Nascimento (2001) studied *Anastrepha obliqua* (Macq.) emergence and mortality factors during the fruiting season of *Spondias dulcis* (Anacardiaceae). The author verified that, out of 1,204 puparia, 53% eclosed, being 48.5%

fruit flies and 4.5% parasitoids. The total of 21.7% of pupae were killed by other mortality agents and 25.3% went through a dormancy period, being 17.8% fruit flies and 7.5% parasitoids. The initial parasitism was 8.6% increased to 15.5% when emergence data of dormant puparia were computed.

In the present work, from 42.4% of puparia neither fruit flies nor parasitoids emerged. This index is similar to that reported by Bressan-Nascimento (2001), being diapause observed exclusively in parasitoids. Therefore, the common practice of discarding uneclosed fruit fly puparia leads to an underestimated parasitism rate.

Diapause involves a marked interruption in insect development. Its induction, maintenance and ending is mediated by biotic and abiotic factors, such as temperature (Masaki & Sakai 1965, Sullivan & Wallace 1968); photoperiod (Masaki 1958, Sullivan & Wallace, 1965); humidity (Tauber & Tauber 1976); food quality and availability (Wallace 1970); host physiological status and population density (Chapman *et al.* 1955, Stewart *et al.* 1967) and intraspecific variation (Matsumoto *et al.* 1951 e 1952, Poitout & Bues 1977).

In the present work, only a proportion of the parasitoid populations entered diapause. Considering also that parasitoid species were submitted to the same biotic and abiotic conditions in the region of the Recôncavo Baiano, then genetic components (intraspecific variation) may be playing a role in the onset of diapause in tropical environments. Thus, the parcel of the population that enters diapause insures temporal escape through unfavorable periods and species persistence until a more favorable season.

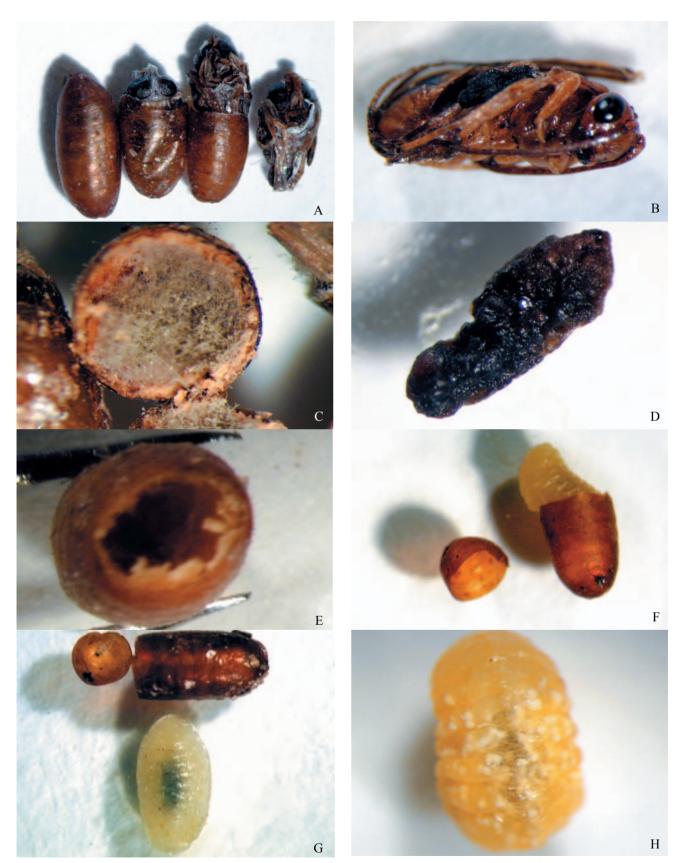


Figure 2. Factors that may lead to non eclosion of fruit fly puparia: (A and B) dehydration, (C) fungus, (D) bacteria, (E) empty puparium and (F, G and H) diapause (n = 752).

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