Growth and Development of Bactrocera Papayae (Drew & Hancock) Feeding on Guava Fruits

M.A.Z. Mohd Noor, A. Nur Azura & R. Muhamad

Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, MALAYSIA

Abstract: Asian Papaya Fruit Fly, B. papayae (Drew & Hancock) undergoes four stages (egg, larva, pupa and adult) during its growth and development. In this study, observation showed that the egg’s stage took 1.16 ± 0.00, larva’s stage took 12.02 ± 0.13 and pupa’s stage took 7.03 ± 0.08 days before the emergence of the adults. The male adult survived for 21.97 ± 2.69 days whilst the female 19.19 ± 1.50 days. It was observed that the eggs laid in a cluster, range between 10 – 50 eggs per cluster. The length and width of the individual egg observed were 1.12 ± 0.03 mm and 0.20 ± 0.00 mm respectively. The percentages of the survived individual larva decreased from the first instar until third instar. In the observation, the length and width of the larva reached 7.77 ± 0.08 mm and 1.84 ± 0.03 mm respectively. Pupae were observed changing in colour from pale yellow to dark brown. The length and the width of the pupae observed were 6.78 ± 0.16 mm and 2.90 ± 0.02 mm. The longevity of the adult B. papayae (Drew & Hancock) was influenced by the diets they consumed, the presence of other individuals, wideness of the areas, differences in times taken in different stages and temperature in the laboratory.

Key words: B. papayae (Drew & Hancock), egg stage, larva stage, pupa stage and adult stage

INTRODUCTION

Asian Papaya Fruit Fly, B. papayae (Drew & Hancock) has been reported infesting 209 hosts across 51 plant families including guava, P. guajava, Linn. (Chua, 1991; White & Elson-Harris, 1992; Clarke et al., 2005; Aluja & Mangan, 2008 and Rwomushana et al., 2008). Infestation of the adults and larvae directly reduces the quantity and quality of the guava fruit. Several intensive studies on the taxonomic and genetic variation of the adults and larvae have been conducted in Malaysia since 1986 (Elson-Harris, 1988; Ooi, 1988 and Vijayasegaran & Mohd., 1991). However, the studies on the growth and development of B. papayae on guava are still lacking. Understanding the growth and development aspects of this insect is important in predicting its development, emergence, distribution and abundance in the field. Due to the reason, this study was conducted with the objective to obtain information on the growth and development of B. papayae feeding on guava fruits.

MATERIALS AND METHODS

Insect rearing:

Colonies of B. papayae were reared by using techniques adopted and modified from Chua (1991), Vargas et al. (2000), Kaspi et al. (2001), Carey et al. (2005), Hee and Tan (2006), Chuang and Hou, (2008) and Wang et al. (2009). Fifty rotten pink guava fruits, P. guajava, Linn var beaumont were collected randomly from the farm belonged to Golden Hope and Beverages Sdn. Bhd., Sitiawan, Perak, Malaysia (N 4° 20’ E 100° 50’). Rearing of B. papayae was conducted under laboratory conditions at 23.92 ± 0.16 C (Min: 21 C; Max: 29 C) and 61.14 ± 0.33% (Min: 51%; Max: 70%) relative humidity (RH) at the Entomology Laboratory, Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia (N 3° 2’ E 101° 43’). Each infested fruit was kept individually in 24.5 x 13.5 x 13.0 cm plastic containers lined with 4.0 cm thick of sterilized vermiculite until the emergence of the adults. Emerged adults were collected and placed into 30.0 x 30.0 x 30.0 cm rearing cage lined with 4.0 cm thick of sterilized vermiculite. Two ends of tissue were soaked in guava juice in a container perforated in two places to allow the tissue to pass through. The flies feed
by sucking the juice from the middle part of the tissue. A mixture solution of honey and yeast extract in 3 : 1 ratio was prepared (Rattanapun et al., 2009). A piece of tissue was soaked in the solution and placed on the floor of the rearing cage. The diets were changed every two days. Six non-infested guava fruits (approximately 100 – 200 g) placed individually on conical flasks in the cage were introduced to the cage as semi natural egging-devices for eggs laying. The egging-devices were kept for five days. These infested fruits were then removed into 24.5 x 13.5 x 13.0 cm plastic containers lined with 4.0 cm thick of sterilized vermiculite to avoid contamination of microbes in the rearing cage. After one week, pupae found in the vermiculite were collected daily by sieving the vermiculite (Somta et al., 2010). Collected pupae were placed back into the cage prepared earlier and kept until the emergence of the adults. Every five months 50 rotten guava fruits were obtained and kept in different cages until the emergence of the adults. These adults were then introduced into the established cage prepared to maintain the wilderness characteristics in the cage colony that used in this study. Death bodies of the adults were removed from the cage every days and feeding devices (food container and tissues) were cleaned every two days to avoid contamination from fungal and bacteria.

The artificial-egging-devices were prepared by adopting techniques established by Chua (1991) and Kaspi et al. (2001). Eggs were collected using a fine brush from the artificial-egging-device which was earlier put in established rearing cage for two hours. The eggs then were soaked into distilled water to determine the viability of the eggs (Vargas et al., 2000). The sank eggs were viable while the floated eggs were unviable. All the viable eggs were placed on black fine mesh (soaked in guava juice earlier) and kept in 90.0 mm diameter petri dishes. The petri dishes were sealed with parafilm to avoid larvae moving out of the dishes. After 24 hours, the petri dishes were observed and first instar larvae were collected and reared in the laboratory condition until the last larva moult (Godin et al., 2002). Ten larvae were taken out daily and they were dipped into hot water (± 95 C) for one minute. Then they were put on tissue paper for drying for two minutes before their bodies’ morphometric measurement (Plate 1.1 and 1.2) were taken. The remaining larvae were then put on 5.0 g diced pulp of the guava individually, kept in 50.0 mm diameter petri dishes. The media (5.0 g guava pulp) that provided to the larvae were changed daily. This process was repeated daily until the last larva moulted. The larvae that formed pupae were transfered to 3.0 x 3.0 cm small vials closed with fine muslin cloth tightened with rubber band. The pupae were kept individually until the adults emergence. The males and females were kept separately in small container sized 15.0 x 20.0 x 10.0 cm covered with fine muslin cloth. The adults were also kept separately according to day when they were emerged. All the adults were fed and supplemented with guava juice, honey and extracted yeast. All the dead individuals in every stages were removed to avoid contamination. The parameters recorded were as follows;

i. durations taken in all stages
ii. survived individuals in each stage
iii. male and female longevity
iv. length and width in each stage excluding the adult’s stage (morphometric parameters)

The morphometric parameters measured was only for the eggs, larvae and pupae which were represented by the means of 40 individual eggs, 40 individual larvae from the first, second and third instars and 40 individual pupae. Other parameters such as sex and change in colour of the adults were also recorded.

Statistical analysis:
The comparison of longevity between the adult male and female was subjected to independent sample t-test performed by SPSS software (version 18).

RESULTS AND DISCUSSIONS

Table 1 indicated four stages of B. papayae during their life, number of individuals, percentages survived and the means of durations taken when the growth and development study was conducted.

<table>
<thead>
<tr>
<th>Stage</th>
<th>No. survival(s)</th>
<th>Survived (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collected eggs</td>
<td>504</td>
<td>100.00</td>
</tr>
<tr>
<td>Egg</td>
<td>484</td>
<td>96.03</td>
</tr>
<tr>
<td>Larva 1st instar</td>
<td>373</td>
<td>74.01</td>
</tr>
<tr>
<td>2nd instar</td>
<td>277</td>
<td>54.96</td>
</tr>
<tr>
<td>3rd instar</td>
<td>133</td>
<td>26.39</td>
</tr>
<tr>
<td>Pupa</td>
<td>87</td>
<td>17.26</td>
</tr>
<tr>
<td>Adult Male</td>
<td>34</td>
<td>6.75</td>
</tr>
<tr>
<td>Female</td>
<td>32</td>
<td>6.35</td>
</tr>
</tbody>
</table>

Table 1: Number of percentage (%) survival during the growth and development of B. papayae (Drew & Hancock)
Egg:

It was observed 96.03% of eggs hatched after 1.16 ± 0.00 days under laboratory condition. The length and width of the egg were 1.12 ± 0.03 mm and 0.20 ± 0.00 mm respectively. The eggs were transparent in colour, cylindrical and tapers gently towards a narrower posterior end, banana shaped as shown in Figure 1a (White & Elson-Harris, 1992; Headrick & Goeden, 1998 and Pena et al., 1998). It was observed that the eggs laid in a cluster form ranging 10 – 50 eggs per cluster (Figure 1b) even in artificially made egging-device once the oviposition took place (Pena et al., 1998). According to Pena et al. (1998), females B. dorsalis (Hendel) and Anastrepha fraterculus (Wiedemann) lay around 1200 – 1500 and 200 – 400 eggs respectively for their entire life in mango. It did not differ much for the numbers of eggs laid even in natural hosts or artificial-egging-devices as the resources provided were sufficient enough for the larvae growth.

Observation showed that there was a different in hatchability durations from B. papayae as compared to other tephritids studied in the laboratory before. Results showed that the eggs of B. papayae hatched earlier (1.16 ± 0.00 days) than B. cacuminata (Raghu, 2002 and Dhillon et al., 2005). Raghu (2002) and Dhillon et al., (2005) reported that B. cacuminata eggs hatched after 42 hours at 25 C and the durations to hatch was between 1.0 day to 5.1 days. The reasons are due to the different host types (pumpkin, bitter gourd, squash gourd, sponge gourd and cucumber), surrounding temperature where the studies were conducted and the species compared (Pena et al., 1998; Raghu, 2002 and Dhillon et al., 2005). Pena et al. (1998) reported that the egg’s stage of fruit flies last from 2 – 20 days.

There were 3.97% of eggs failed to hatch and this was related to the temperature fluctuation in the laboratory even though it was under a controlled environment. Increment or decrement in temperature may affect the viability of the eggs. Golizadeh et al. (2009) reported that when temperature exceeds the tolerant limit of hatchability, the eggs of the insects will not hatch. In this study, the hatchability of B. papayae eggs was observed at 23.92 ± 0.16 C and this temperature was suitable condition for eggs to hatch.

Larva:

Bactrocera papayae underwent three larval instars (Figure 2). According to Chang et al. (2007), a large majority of the larvae often died after reaching the third instar. Result obtained showed that the B. papayae larval survivorship decreased as the times passed from one instar to another. The percentage of survived larvae decreased from 74.01% (first instar) to 54.96% (second instar). Even though the percentage of eggs hatched was high (96.03%), only a few (26.39%) of the larvae succeeded in reaching the third instar stage due to the microbial infestation of the eggs or on the guava pulp which was transmitted when the diets were changed and exposed to the laboratory environment (Chang et al., 2007).

Fig. 1a: Single egg of B. papayae (Scale: 0.16 mm); b) a cluster of B. papayae eggs (Scale: 0.42 mm)

Fig. 2.a: 1st instar larva; b) 2nd instar larva; c) 3rd instar larva
The larvae survived for 12.02 ± 0.13 days before pupation took place and this period taken was shorter compared to what have been described by Pena et al. (1998) where the larval stage of fruit flies was between 2 – 4 weeks. Within this duration, it was observed that the larvae reached 7.77 ± 0.08 mm length and 1.84 ± 0.03 mm width. According to Pena et al. (1998), full grown larvae measures approximately 7.00 mm in length but they did not mention the body width. White and Elson-Harris (1992) reported that third instar larva of the fruit flies average size is 6.50 – 10.00 mm in length 1.00 – 1.50 mm in width. Only 17.26% larvae survived to pupation.

**Pupa:**

Pupation starts from the prepuparial stage in which the mouthparts are invaginated and the integument takes on a waxy appearance (Headrick & Goeden, 1998). The duration of the prepupa within the puparium is unknown. The prepupal integument is shed and adheres to the innerwall of the puparium. The pupa forms within the puparium after the prepupal moult.

During the observation, the pupae took 7.03 ± 0.08 days before the adult emerged. The duration taken in this stage was shorter than the other species which generally took 2 – 4 weeks. Different in durations taken to form pupa mainly due to temperatures and relative humidity. Stresses due to environmental changes in most cases hasten the growth of the insects for survival (White and Elson-Harris, 1992 and Pena et al., 1998).

On the average, the pupae sizes were 6.78 ± 0.16 mm in length and 2.90 ± 0.02 mm in width. The colour of pupae gradually changed from pale yellow to dark brown as the times changed for pupae to develop (Figure 2; from left to right). According to Headrick and Goeden (1998), the processes of hardening and darkening of the integument during the pupae development are within certain time frame. The pupa’s shape was oval or exarate (White & Elson-Harris, 1992). In natural environment or in the field, mature larvae of *B. papayae* drop to the ground and move into the soil and find preferable pupariation site which is usually underneath the plant host (White & Elson-Harris, 1992 and Pena et al., 1998).

**Adult:**

Figure 4 shows the adult male and female of *B. papayae*. Adults emerged after eight days of pupation at temperatures of 23.92 ± 0.16°C. The emergence of the *B. papayae* (Drew & Hancock) adults in this study was faster compared to *B. cacuminata* (Hering) as described by Raghu (2002). He reported that, at 25 C, the pupae of *B. cacuminata* (Hering) took approximately 12 days before the emergence of the adults.

Morphologically, according to White and Elson-Harris (1992), the scutum of adult’ *B. papayae* predominantly black with lateral yellow stripes, a black T-shaped mark on both males and females abdomen and typical dacine wing pattern. The males posses pecten. There were yellow marks on the thorax and made the *B. papayae* wasp-like appearances (Fletcher, 1987).

---

**Fig. 3:** Changes in colouration of pupa (from left to right); Scale: Bar 2.20 mm

**Fig. 4:** Adult of *Bactrocera papayae*
In this study, it was observed that the longevity of the male was 21.97 ± 2.69 days while female was 19.19 ± 1.50 days. Statistically, the longevity of the male was observed not significantly (P>0.05) longer than the female. There were no indication as to which sex might survive longer compared to the other and yet still can be discussed since the longevity are influenced by a vast of factors.

The growth of fruit flies and longevity of the adults tephritids depends on the diet consumed (Vargas et al., 2000 and Zur et al., 2009). The diets, either natural, semi-natural or artificially made, which are provided during the rearing may contribute to longevity periods. In this study, concentrated honey as sugar sources enriched with carbohydrates and concentrated yeast extract as protein sources were provided for the adults. Besides protein, concentrated yeast extract also provides the vitamins and minerals needed by the adults. The nutrients in the diets play important role and their functions is crucial for insects to grow and develop. For instant, carbohydrate provides energy for routine life activities such as flight (Zur et al., 2009 and Wang et al., 2009).

Viable source will extend the longevity but if the source provided early, the flies will utilize it, reproduced and died earlier (Wang et al., 2009). In contrast, Canato and Zucoloto (1997) stated that sugars source were important for the C. capitata female’s adult since the insect successfully producing eggs without ingesting protein. Whilst, Tsiropoulos (1977), stated that some of the Rhagoletis species such as R. completa Cresson, R. pomonella (Walsh) and R. cingulata (Loew) can survive and able to produce eggs on carbohydrate and water alone.

Other factors which affect the growth, development and longevity were the presence of other individuals in the surrounding areas. In the study, males and females were kept separately, therefore the males and females could only lived for 21.97 ± 2.69 days and 19.19 ± 1.50 days respectively. With the presence of other individuals either same or different sex in the optimum density, the flies may potentially survived longer than the results revealed and this was reported by Meksomngsee et al. (1988) where B. tau can survived up to 148 days. Dhillon et al. (2005) reported that the longevity of B. cucurbitae can last from 21 – 179 days.

The area of the fly influence the growth, development and longevity where the flies can mobilize to fulfill their requirements and needs (Zur et al., 2009). Theflies need shelter, foraging for foods, find mates and other life routines for their survival. Big areas usually provide all the requirements of the flies. However, if the area is big but too dense it may affect the life of the flies. In this study, B. papayae adults individually kept in the small container sized 15.0 x 20.0 x 10.0 cm covered with fine muslin cloth. Even though, the containers were not dense with flies, food and supplements but the areas probably not enough for the flies to move freely. Polyphagous and multivoltine tephritids are known for their high mobility thus the distribution wide across the region (White & Elson-Harris, 1992).

The different time taken for each stage from the egg to adult may also affect the life cycle and the longevity (Pena et al., 1998). Different stages face and experience different needs. For example, larvae need to feed more during growth stage so that they can develop well to adulthood. Fernandes-Da-Silva and Zucoloto (1993) reported that C. capitata larvae utilized the nutrients of the oranges mainly from the lower part of the fruits where the nutrients are denser. They also found that the longevity was shorter when compared to papaya but higher in the emergence of the adults.

The longevity of B. papayae in this study was also influenced by the temperature inside the laboratory. Mean temperature in the laboratory was 23.92 ± 0.16°C and fluctuated with very minimal changes. There were reports stated that temperature was very crucial in the life cycle longevity of the fruit flies (Tsiropoulos, 1977; Pena et al., 1998; Vargas et al., 2000; Dhillon et al., 2005; Golizadeh et al., 2009 and Nyamukondiwa & Terblanche, 2009). The temperature was also reported influenced the maturation and sexual behaviour of the males Anastrepha ludens, Anastrepha obliqua, Anastrepha serpentina and Anastrepha striata (Aluja & Mangan, 2008). According to Vargas et al. (2000), both adults male and female of B. cucurbitae, B. dorsalis (Hendel), C. capitata survived in different durations at different temperature. Golizadeh et al. (2009) reported that temperature affected the specific rate functions of survival, reproduction, population growth and development of many insects and this directly influence the life cycle and the longevity.

**Conclusion:**

_Bactrocera papayae_ is a relatively less studied species in Malaysia specifically on the growth, development and longevity. As the other tephritid fruit flies, _B. papayae_ underwent four stages namely egg, larva, pupa and adult in their life. The eggs, larvae and pupae took 1.16 ± 0.00, 12.02 ± 0.13 and 7.03 ± 0.08 days respectively. _Bactrocera papayae_ completed all the stages within 20.21 ± 0.21 days. Statistically there was no different of the longevity between the male (21.97 ± 2.69 days) and the female (19.19 ± 1.50 days). The
longevity are influenced by a few of factors such as the different stages within the life cycle, laboratory temperature and relative humidity, guava fresh dice for larvae consumptions and supplements provided for the adults.

ACKNOWLEDGMENTS

This research was funded by MOA Grant no. 05 – 01 – 04 – SF 1018. The authors would like to express their gratitudes to Mr.Md. Roslan Zulkifli (Senior Manager), Golden Hope Food & Beverages Sdn. Bhd. and his staffs for their help in conducting the sampling. Many thanks to Mrs. Suhana binti Yusof (Pest and Disease Management Programme, Horticulture Research Centre, MARDI Headquaters, 43400 Serdang, Selangor,MALAYSIA) for her advise in rearing technique of fruit fly.

REFERENCES


