

Life Table of *Cochlochila bullita* Stål (Hemiptera: Tingidae) on *Orthosiphon aristatus* (Blume) Miq. and *Ocimum basilicum* L. in Laboratory Conditions

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ABSTRACT

Ocimum tingid, *Cochlochila bullita* Stål (Hemiptera: Tingidae) is a pest of Lamiaceae plants such as basil, tulsi and coleus. It is now being recorded in Malaysia as a pest of the cat's whiskers plant, *Orthosiphon aristatus* (Blume) Miq. Nevertheless, apart from its brief biological description, no other information is available. The life table of this pest was studied in laboratory conditions. Development time for *C. bullita* feeding on *O. aristatus* was 23.3 ± 0.9 days, which was found to be similar to those feeding on *Ocimum basilicum* (22.8 ± 0.3 days). Although *C. bullita* posts a higher mortality rate on *O. aristatus* than on *O. basilicum* (52% vs. 37%), the adult longevity of the bugs that feed on *O. aristatus* (♀: 33.9; ♂: 38.2 days) was found to be significantly higher than those bugs that feed on *O. basilicum* (♀: 27.2; ♂: 26.0 days). The pre-oviposition, oviposition and fecundity of *C. bullita* were also different between the host plants. The net reproductive rates (R_0), finite rate of increase (λ) and intrinsic rate of increase (r) were also higher on *O. aristatus* (10.7504, 1.0690 and 0.0667), although there was an increased in immature survival on *O. basilicum* (6.0287, 1.0556 and 0.0541). Therefore, it is concluded that *O. aristatus* is

as good as *O. basilicum*, or the population growth of *C. bullita* is more favoured as compared to *O. basilicum*.

Keywords: Basil, cat's whiskers plant, lace bug, life table parameter, Malaysia, *Ocimum tingid*

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INTRODUCTION

Ocimum tingid, *Cochlochila bullita* Stål (Hemiptera: Tingidae) is a lace bug associated with the plants of the Lamiaceae family. The lace bug has been recorded to attack culinary herbs, sweet basil, *Ocimum basilicum* L. and *Ocimum tenuiflorum* L. in China, India and also some parts of South East Asia (Livingstone & Yacoob, 1987). In Malaysia, the lace bug was first recorded in Selangor, attacking a native plant called *Orthosiphon aristatus* (Blume) Miq., which is a medicinal plant with many health benefits (Sajap & Peng, 2010). In a recent survey, *C. bullita* was found to attack *O. basilicum* planted in Universiti Putra Malaysia. The lace bugs, nymphs and adults fed on the leaves and young shoots of the herb. The infested leaves and shoots became wilted, dried and in many cases, the whole plant died upon heavy or repeated infestations. Meanwhile, pattern of the infestations showed that the lace bugs could pose a serious threat to the herbs and their related industries if they were to be planted in a wider scale. Apart from the life history of *O. basilicum* that has been documented in Thailand (Tigvattnanont, 1989) and related papers by Sajap and Peng (2010) and Peng *et al.* (2013), very little information is available on this particular insect. Even though *C. bullita* appears to be thriving on both plants, its performance in terms of its demographic characteristics on both plants has yet to be known. Demographic parameters of an insect can be elucidated using a life table analysis (Carey, 1993; Southwood & Henderson, 2000). These

demographic parameters can be used to estimate the insect's population growth capacity on different regimes such as host plants (Greenberg *et al.*, 2001), prey for predatory insects (Legaspi *et al.*, 2008), temperature and strain (Liu & Meng, 1999; Bong *et al.*, 2012). The parameters in the life table, particularly the intrinsic rate of natural increase (r), can be used to evaluate the magnitude of suitability of a host plant to an insect (Razmjou *et al.*, 2006). Plants that render lower values of r are relatively more resistant or less suitable hosts than those with higher values of r . In this experiment, life table was used to compare the performance of the lace bug *C. bullita* on its hosts, *O. aristatus* and *O. basilicum*. Understanding the growth and development performances of the lace bug on its host plants will lead to development of a better management strategy of the pest.

MATERIALS AND METHODS

The experiments were conducted in the Entomology Laboratory at the Faculty of Forestry in a room at 28 ± 2 °C, $55 \pm 5\%$ RH, and 12h:12h (L:D) photoperiod.

Rearing Method

The lace bugs were reared on potted plants of *O. aristatus* and *O. basilicum*, which were kept in cages (60 × 30.5 × 25.5 cm). Ten pairs of its females and males were introduced into the cages and allowed to breed on the plants. The plants were regularly watered and replaced with new potted plants when wilted. In order to eliminate residual effects of the previous host plant, *O. aristatus* or

the lace bugs intended for the experiment on *O. basilicum* were collected from the F2 generations of the lace bugs that had been reared on *O. basilicum*.

Preparation of the Host Plants

Stems of *O. aristatus* and *O. basilicum* were cut to about 8 cm long and inserted into a plastic cup (3.5 cm diameter × 4 cm height) filled with water through a hole drilled in the lid. The rooted stems (about two weeks old) were used for the experiments.

Life Table Study

Thirty pairs of the females and males (around 1 – 3 days old) were collected from the colonies and placed on a Petri dish (Brandon Disposable Petri dish; 9.0 × 1.5 cm) containing a piece of moistened filter paper (Advantec Grade No. 1 Qualitative Filter Paper; Diameter: 5.5 cm) and two leaves of the respective plants that served for their food and mating arena. The lace bugs were kept in pairs for 7 d and gravid females were taken for oviposition. For oviposition, three rooted stems of *O. aristatus* or *O. basilicum*, as prepared earlier, were placed separately in a plastic container (MS Venture C1-MS30; 11.8 × 10.2 cm) with its lid covered with muslin cloth. Ten gravid females were placed separately in the container for oviposition. After 24 h, the females were removed and transferred into new containers with fresh plants. The eggs were counted, kept on the plants till hatched and their incubation period was also recorded. Once the eggs

hatched, the neonates were counted and left on the plants till moulted. The second instars were then transferred individually into plastic cups (MS Venture C5-MS4; 7.5 × 3.9 cm) containing a moistened filter paper and a fresh leaf. The cups were covered with perforated lids. The leaves and moistened filter paper were changed every 2 d. The nymphs were examined daily and their moulting and survival were also recorded. Successful moulting from each stadium was confirmed with the presence of exuviae. A cohort of 100 eggs was used for each of three replicates. Thus, a total of 300 eggs were used for each host plant in the life table experiment. The fecundity, reproductive period and adult longevity of the lace bugs were studied using the adults collected from the development and survivorship experiments. The adults were collected within 24 h after emergence. They were sexed and the sex ratio was recorded. They were then randomly paired and kept in plastic cups. A total of 30 pairs for a cohort were used for this experiment. If the male died earlier, it was replaced with a new male to ensure continuity of mating and oviposition. The pairs were kept until death. Longevity of adults, pre-oviposition, oviposition, post-oviposition periods and fecundity were recorded. Non-laying females were excluded from the data analyses. The possibility of reproduction without mating was also investigated. These virgin females were kept separately in plastic cups and examined every 24 h for oviposition.

Experimental Design and Statistical Analysis

The experimental design for constructing the life table was modified from the design in the life history of *Maconellicoccus hirsutus* (Chong *et al.*, 2008). All life table and fertility table parameters were measured and calculated as described by Birch (1948). Population parameters were calculated as:

x : Age classification

l_x : Probability of surviving to age x

m_x : age-specific fecundity, the number of female eggs born per female in each interval class

GRR : Gross reproductive rate, $GRR = \sum m_x$

Intrinsic rate of increase (r) is estimated using the Euler-Lotka equation with age indexed from zero, $\sum e^{-r(x+1)} l_x m_x = 1$, $x = 0$ to 40

R_0 : Net reproductive rate, $R_0 = \sum (l_x m_x)$

T_G : Mean generation time,

$$T_G = (\ln R_0) / r.$$

λ : Finite rate of increase, $\lambda = e^r$

DT : Doubling time, $DT = \ln 2 / r$

RESULTS

Development Time and Survival

Eggs of *C. bullita*, which were held at 28 ± 2 °C, hatched in 8.8 ± 0.2 d. When fed on *O. aristatus*, the lace bug had a cumulative developmental time from egg laid till adult emergence ranging from 19.8–28.3 d, with an average of 23.3 ± 0.9 d. All the nymphs underwent five moults. The lace bugs had a relatively uniform and shorter development time on *O. basilicum* ranging from 21.4 to 24.4 d, with an average of 22.8 ± 0.3 d.

Although the development time of the third instars was longer on *O. aristatus* and so did the fifth instars on *O. basilicum*, there was no significant difference on the overall development time of *C. bullita* immatures feeding on both the host plants ($t_{12} = 0.0086$; $P = 0.589$). They lived significantly longer on *O. aristatus* than those on *O. basilicum* (Table 1). The longevity of the males and females on the same host plant, however, was not significantly different. The host plants also influenced the mortality of the immatures. Significant differences in mortality occurred in the early nymphal stages. The mortality of the first instars on *O. aristatus* was about 10% higher compared with that on *O. basilicum* ($t_{18} = 2.626$; $P = 0.017$). Meanwhile, the second instars had a mortality of 7.3% on *O. aristatus* and 3.0% on *O. basilicum* ($t_{18} = 2.847$; $P = 0.010$). These differences contributed to the overall mortality from the egg to adult stages of 52.0% on *O. aristatus*, which was significantly higher compared to 37.0% on *O. basilicum* ($t_{18} = 2.518$; $P = 0.022$) (Table 2). The genders of the pre-adult mortality were assumed using the male to female ratio of *O. aristatus* and *O. basilicum*, which was 2:3 and 1:1, respectively. In general, the survival rate (l_x) on both plants followed almost a similar pattern with a higher mortality occurred during the nymphal stages, especially during the first instar and this decreased gradually as they matured, following a type III survivorship curve, as described by Speight *et al.* (1999) (Figures 1 and 2).

TABLE 1
Development times of *C. bullita* immature stages feeding on either *O. aristatus* or *O. basilicum* leaves under laboratory conditions

Stage	Development time (Days \pm SE)					
	<i>n</i>	<i>O. aristatus</i>	<i>n</i>	<i>O. basilicum</i>	<i>t</i>	<i>P</i>
Egg	300	8.8 \pm 0.2 a	300	8.8 \pm 0.2 a	0.034	0.973
1st instar	277	2.8 \pm 0.3 a	273	2.4 \pm 0.1 a	1.486	0.154
2nd instar	220	2.4 \pm 0.1 a	244	2.0 \pm 0.1 a	1.658	0.115
3rd instar	198	2.4 \pm 0.1 a	235	2.1 \pm 0.1 b	2.659	0.015
4th instar	188	2.8 \pm 0.2 a	230	2.7 \pm 0.1 a	0.232	0.819
5th instar	176	4.0 \pm 0.2 a	217	4.7 \pm 0.2 b	2.151	0.045
Egg - Adult	144	23.3 \pm 0.9 a	189	22.8 \pm 0.3 a	0.551	0.589
1st - Adult	144	14.5 \pm 0.8 a	189	14.0 \pm 0.2 a	1.196	0.247
Longevity (Females)	60	33.9 \pm 2.8 a	48	27.2 \pm 1.7 b	2.050	0.049
Longevity (Males)	78	38.2 \pm 3.3 a	69	26.0 \pm 2.3 b	3.014	0.004

Means followed by different letters in the row are significantly different at $P = 0.05$ (t-test; SPSS)

TABLE 2
Mortality rates of *C. bullita* immature stages feeding on either *O. aristatus* or *O. basilicum* leaves under laboratory conditions

Stage	Mortality (% \pm SE)			
	<i>O. aristatus</i>	<i>O. basilicum</i>	<i>t</i>	<i>P</i>
Egg	7.7 \pm 3.0a	9.0 \pm 2.9a	0.309	0.761
1st instar	19.0 \pm 3.1a	9.7 \pm 1.9b	2.626	0.017
2nd instar	7.3 \pm 1.3a	3.0 \pm 0.8b	2.874	0.010
3rd instar	3.3 \pm 1.0a	1.7 \pm 0.6a	1.510	0.148
4th instar	4.0 \pm 1.6a	4.3 \pm 1.0a	0.005	0.960
5th instar	10.7 \pm 2.8a	9.3 \pm 1.8a	0.377	0.710
Egg - Adult	52.0 \pm 4.9a	37.0 \pm 2.9b	2.518	0.022

Means followed by different letters in the row are significantly different at $P = 0.05$ (t-test; SPSS)

Fecundity

The age-specific fecundity (m_x) and duration of oviposition varied correspondingly with the host plants they fed on (Fig.1 and Fig.2). The lace bugs feeding on *O. aristatus* began to oviposit 1 d earlier and this lasted 7 d later than those feeding on *O. basilicum*. Their fecundity peaked on the 9th and

11th day on *O. basilicum* and *O. aristatus*, respectively, after adult emergence. With a longer oviposition period, the lace bugs feeding on *O. aristatus* had a higher mean of fecundity than on *O. basilicum*. As shown in Table 3, the lace bugs had a significantly shorter pre-oviposition ($t_{38} = 2.404$; $P = 0.021$), longer oviposition period ($t_{38} =$

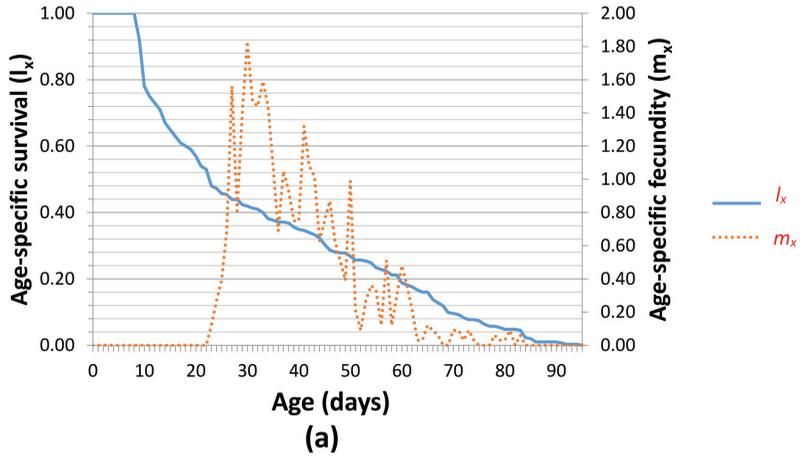


Fig.1 Daily age-specific survival (l_x) and fecundity (m_x) of female *C. bullita* feeding on *O. aristatus*

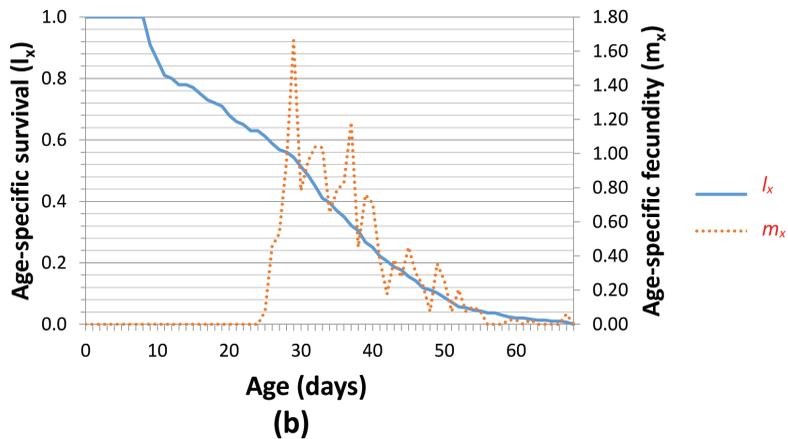


Fig.2: Daily age-specific survival (l_x) and fecundity (m_x) of female *C. bullita* feeding on *O. basilicum*

2.154; $P = 0.0138$) and higher fecundity ($t_{38} = 3.090$; $P = 0.004$) on *O. aristatus* than those on *O. basilicum*. Meanwhile, the male to female ratio on *O. aristatus* and *O. basilicum* was 2: 3 and 1:1, respectively.

Life Table Parameters

The population parameters are summarised in Table 4. The intrinsic rate of natural increase (r) of the lace bug on *O. aristatus*

was 0.0667 per female per day and the daily finite rate of increase (λ) was 1.0690 female progenies per female per day, with a mean generation time (T_c) of 35.59 days. The population doubled its number in 10.39 d. The gross (GRR) and net (R_0) reproductive rates were 30.75 and 9.47, respectively. On the other hand, the gross (GRR) and net (R_0) reproductive rates of the bug on *O. basilicum*, were 16.41 and 6.03,

TABLE 3
Reproductive parameters of *C. bullita* feeding on *O. aristatus* and *O. basilicum*

Parameters	<i>O. aristatus</i>	<i>O. basilicum</i>	t	P
Pre-oviposition (days)	5.1 ± 0.4 a	6.3 ± 0.3 b	2.404	0.021
Oviposition (days)	24.3 ± 2.5 a	17.7 ± 1.8 b	2.154	0.038
Post-oviposition (days)	3.6 ± 0.8 a	2.5 ± 0.7 a	1.054	0.298
Fecundity (eggs)	51.3 ± 4.5 a	32.4 ± 4.1 b	3.090	0.004
Sex ratio (female, %)	59.7 ± 5.4 a	50.3 ± 5.3 a	1.250	0.227

Means followed by different letters in the row are significantly different at $P = 0.05$ (t -test; SPSS)

TABLE 4
Population parameters of *C. bullita* feeding on *O. aristatus* or *O. basilicum*

Parameter	<i>O. aristatus</i>	<i>O. basilicum</i>
Gross reproductive rate (GRR)	30.75 ± 2.6910 a	16.41 ± 2.0373 b
Net reproductive rate (R_0)	10.75 ± 6.84 a	6.03 ± 4.45 b
Mean generation time (T_G)	35.59 ± 11.22 a	26.92 ± 10.05 b
Intrinsic rate of increase (r)	0.0667 ± 0.0293 a	0.0541 ± 0.0227 b
Finite rate of increase (λ)	1.0690 ± 0.3208 a	1.0556 ± 0.3171 b
Doubling time (DT)	10.39 ± 5.24 a	12.80 ± 5.42 b

Means followed by different letters in a row are significantly different at $P = 0.05$ (t -test; SPSS)

respectively. The population doubled its number in 12.80 days. The intrinsic rate of natural increase (r) was 0.0541 per female per day, whereas the daily finite rate of increase (λ) was 1.0556 female progenies per female per day, with a mean generation time (T_G) of 26.92 days.

DISCUSSION

There are many factors influencing growth, longevity and reproduction of an insect in its natural environment. One of the factors that has been shown to greatly influence development of an insect is food quality (Ellers-Kirk & Fleischer, 2006). This factor was also found to have affected *C. bullita* feeding on two different host plants. Even

though immatures of *C. bullita* feeding on both plants had a similar development time, they differed significantly in their survival and fecundity. The lace bugs feeding on *O. aristatus* had a lower survival rate in the early stages of development but with a longer longevity with a higher fecundity than those feeding on *O. basilicum*. These differences indicated that *O. aristatus* could be a nutritionally better host than *O. basilicum*. Such an effect has been widely reported to occur on many herbivorous insects (Awmack & Leather, 2002). Even though *O. aristatus* could be nutritionally better, the leaves and stems are phenotypically covered with relatively dense and long trichomes that could have physically hampered young

nymphs from accessing to the leaf surfaces and led to starving individuals. This physical defence mechanism by plants against insect herbivores occurs on wheat against bird cherry oat aphids (Roberts & Foster, 1983) and potatoes against potato leafhopper (Medetros *et al.*, 2005). Like those sap sucking insects, the surviving older lace bug nymphs were able to thrive well on the plant as their longer stylet could have easily reached the leaf surfaces to suck the sap.

Even though, the lace bugs in Malaysia were found to be able to develop and reproduce successfully on *O. basilicum*, their rates of development and reproduction are different from that of the lace bugs feeding on *O. basilicum* in Thailand (Tigvatnont, 1989). The Malaysian lace bugs have a relatively shorter life span and less fecundity than those lace bugs attacking *O. basilicum* in Thailand. The reasons, apart from the varietal differences of the host plant, could be the climatic differences between the two countries. The warmer temperature of Malaysia, in comparison with the cooler temperature of Thailand, shortens the longevity and reduces the oviposition rate of the lace bugs. These differences are expected as insects usually have a shorter lifespan and less reproductive in the warmer temperature than those insects in the cooler subtropical regions (Chong *et al.*, 2003). The present work is based on the traditional female age-specific life tables which focus only on the survival and the fecundity of the female population. Problem of stage overlapping throughout the age-specific life table might occur and cause

inaccurate estimation. Besides, the improper manipulation of the survival and fecundity curves due to unknown sexes of immature are very likely to occur and lead to error in the derived parameters (Huang & Chi, 2012). Thus, improvisation could be done by performing the age-stage two-sex life tables (Chi 2008) in the future.

CONCLUSION

The lace bug *C. bullita* was found to thrive well on both host plants and has the potential to become a serious pest for the two widely distributed crops. With their shorter lifespan under a relatively warm and constant tropical climate, the insect may have overlapping generations that could contribute to its significance as a pest of *O. aristatus* and *O. basilicum*. The higher rate of increase on *O. aristatus* suggests that *O. aristatus* is as good as *O. basilicum*, or even more, favours the population growth of *C. bullita*. In this study, however, the cohort life table was constructed under a controlled environment that eliminated many natural variables which might have affected the parameters. Thus, the life table data obtained in this study were just an insight into the demographics of *C. bullita* populations to enable us discern patterns and make predictions about the changes of its populations in the future. The insect's ability to adapt into a wide geographical location with different climatic regimes indicates that the insect can be invasive to regions with prevailing host plants. Thus, fundamental knowledge on its biological characteristics is essential to develop a better

integrated pest management strategy against this particular pest.

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