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Research Article

Biology and Demography of *Helopeltis bradyi* Waterhouse (Hemiptera: Miridae) Reared on Cucumbers

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ABSTRACT

Helopeltis bradyi is often used as for research and is required in large numbers. The insects were obtained from cacao plants and reared with cucumber fruit in the laboratory. This study aimed to determine the biology and demography of *H. bradyi* reared on cucumber under laboratory conditions. The demographic parameters observed were net reproduction rate (Ro), gross reproduction rate (GRR), intrinsic growth rate (r), average generation period (T), and population doubling (DT). The results showed that the egg stage lasted for 6.33 ± 0.47 days, the developmental period of the I–V instar nymph was 2.13 ± 0.34, 2.07 ± 0.25, 2.13 ± 0.34, 2.33 ± 0.47, and 3.20 ± 0.40 days. The lifespan of male imagoes were 32.33 ± 4.92 days, and female imagoes were 24.60 ± 6.64 days. The adult female pre-oviposition period was 2.2 ± 0.40 days and oviposition period of 22.9 ± 4.66 days. *H. bradyi* has a type III survival curve which indicated high mortality rates during the first instar egg and nymph population. Demographic statistics of *H. bradyi* on cucumbers fruit are GRR of 296 individuals per generation, Ro of 196 individuals per parent per generation, rate of r of 0.18 individuals per parent per day, T of 29.34 days, and DT of 3.85 days.

Keywords: Cohort; developmental period, laboratory; lifespan; survival curve

INTRODUCTION

Helopeltis spp. is a major polyphagous pest in cacao, tea, guava, and cashew nut plantations. This insect attacks by piercing and sucking fluids from host plant causing damage on attacked plant parts, such as fruit and young shoots. Their attacks result in black brownish lession that then turn into necrotic lesion and plants to dessicate (Saroj & Vanitha, 2015).

Damage of *Helopeltis* decreases quantity and quality of yield in plantations. In Indonesia, damage severity of 65.50% was able to decrease production of Kiaran 8 tea shoot by 87.60% for 8 weeks, resulting in total lost of 40% and prediction loss to range between 50-100% (Sukasman, 1996; Widayat *et al*., 1996). In addition, this insect is a pest of cacao and high infestations of this pest can cause upto 50% yield loss (Indriani, 2004).

In Indonesia, this insect was first known as *H. antonii* (Stonedahl, (1991). Melina *et al.* (2016a) confirmed that the *Helopeltis* species known to attack cacao in Yogyakarta was *H. bradyi*.

Description of insect lifecylecs is essential in crop protection and pest management. Studies regarding insect demography and population dynamic are essential information in insect ecology. Demographic and life table are different, but related aspects of insect lifes. Demographic is an analysis of a populations quantitative characteristics, especially correlations between population growth, resilience, and population mobility, while life tables is an important tool to understand insect population dynamics in a lifecycle. Life tables can be useful approachs in entomology where growth and mortality vary across life stages (Kakde *et al.*, 2014). Life tables are able to summarise survival probabilities and insect population reproduction on different hosts and environmental conditions. Demographic and population dynamic of insect can explain how insect population grow and decrease (Price *et al.*, 2011).

The choice of *H. bradyi* as a research object is due to its status as an important plantation pest; thus, making it required in larger numbers and continuous matters. Basic information of *H. bradyi* is an initial step to determine proper management strategies. Understanding of pest demographic statistics is important to forecast insect population growth and development of pest management strategies (Tsai & Liu, 2000). This study aimed to determine biology and demography of *H. bradyi* reared on cucumbers in lab conditions.

MATERIALS AND METHODS

This study was conducted at the Laboratory of Pest Control Technology, Biological Control Section, Department of Plant Protection, Faculty of Agriculture, Universitas Gadjah Mada, from January to May 2021. Room temperature during this study ranged between 28–30°C with relative humidity of 55–65% without air conditioning.

Helopeltis bradyi Rearing Cohort

As much as 50 *H. bradyi* nymphs were collected from cacao plants at PT Pagilaran Samigaluh plantation, Kulon Progo, Yogyakarta (7°40'27.5"S 110°08'19.7"E). Field collected nymphs were then reared until it produced three generations and was then used for testing. Food sources used for *H. bradyi* rearing was cucumber with the size of 10-15 cm and stored in containers (Figure 1). Three container sizes were used for different insect life stages. For imagoes, the container used had diameter of 11 cm and height of 19 cm, for egg incubation (Figure 2A) had diameters of 14 cm and height of 16 cm, while containers for nymph rearing size (Figure 2B-F) was the same with containers for egg incubation. A pair of male (Figure 2G) and female (Figure 2H) imagoes from the first generations was placed in the rearing containers to induce mating. Biological and demographic observation started from egg pahse until all imagoes died. Eggs obtained from mating processes (Figure 3A) of a *H. bradyi* pair was used as a cohort. Eggs were inserted (Figure 3B) into the tissue of cucumber surfaces, counted and cucumbers with eggs were transferred to egg incubation containers for further rearing until eggs hatched into nymphs. This study used egg cohorts. As much as 262 first instar nymphs were transferred to nymph rearing containers and fed with 2 cucumbers that were positioned standing, each container was filled with maximum 20 nymphs. Cucumbers were changed every two days to maintain fresh food sources. Rearing procedures for the second to last instar were similar with first instar nymphs. The number of surviving, dead, and molted nymphs were observed and development was recorded everyday until reached imago. Molting were indicated by exuvia. Second generation male and female imagoes were reared similarly to the first generation, one pair per container. Containers for *H. bradyi* imago reraing were cleaned everyday to prevent contamination.

Observation parameters

Daily observations included survival of *H. bradyi* cohort since eggs, nymph, until all imagoes died. Duration and percentage of eggs that hatched were observed during the eggs stage. Duration and and death percentage of nymph were observed during the nymph stage. Molting were observed and recorded everyday until nymphs developed to imagoes. For imagoes, sex ratio, daily oviposition, number of dead imagoes, and lifespan of male and female were observed and recorded during this study.

Data Analysis

Biological statistic, life table and demography anlaysis of *H. bradyi* adopted methods from Triwidodo *et al.*(2020). Results from observation were arranged in life tables, including *x* as the age class. The *lx* is the survival chance of an invidual at age *x*. *dx* is the number of individuals that died at each age *x*. *Lx* is the average number of individual that survived between *x* and *x+1*. *qx* is the morality proportion at each age *x*. *Tx* is the total number of individuals at *x* and besides at age *x*. *ex* is the life expectation left for an individual at *x*. *mx* is the fecundity of each individual at age *x* or number of nymphs that hatch at age *x*. *H. bradyi* demographic statistics was analyzed according to Birch (1948). Net reproduction rate (Ro), was calculated using : Ro = $\sum lxmx$. Gross reproduction rate (GRR), was calculated using: GRR = $\sum mx$. Intrinsic growth (r) was calculated using: r = (ln Ro) / T. Average generation period (T) was calculated using: T = $\Sigma\, xlxmx / \Sigma\, lxmx$. Doubling time (DT) was calculated using: DT = ln (2) / r. Data of *lx* and *mx* were plotted according to time to demonstrate survivorship and reproduction of *H. bradyi*. Microsoft Excel 365 was used to analyze data.

RESULTS AND DISCUSSION

***Helopeltis bradyi* Biology**

Sufficient food was provided and *H. bradyi* were protected from natural enemies implying that death during this study occurred due to physiological causes and age. Female imago of *H. bradyi* oviposited their eggs on the surface of cucumber tissue (Figure 3B). Eggs are shown to have a pair of uneven slight threads (Figure 2A). Eggs were placed singularly of in clutches. After hatching, instar I nymphs (Figure 2B) were yellow-oranged and have setae on almost all part of its body. Legs were darker, covered with setae, and have not developed scuttelar spine on thorax. Eyes and last segment of last abdomen was reddish on posterior and darker on lateral.

Second instar nymphs (Figure 2C) had less setae on their bodies compared to the previous stage, but more on their antenae. Body surfaces were orange and slightly darker on the lateral sides of head, thorax, and strips of the abdomen dorsal side. Eyes were dark red, first segment of antennae were orange and the next segment were darker. Scuttelar spine started to be noticeable.

Third instar nymphs (Figure 2D) were dark orange on the lateral side of its their body, legs and first antennae segments were orange with grey stripes on each segment. Setae were not found on their body of antennae. Wings were still short and started to appear dark orang with dark red eyes. Scuttelar spine have developed longer and orange.

Fourth instar nymphs (Figure 2E) were orange on their head, thorax, and posterior side of their abdomen. Leg and first antennae segments were orange with darker grey stripes on other segments. Eyes were dark red, scuttelar spine were orange and observable. Wing buds were brown and grow to cover one third of their abdomen.

Fifth instar nymphs (Figure 2F) is the last nymph instar before adults. Fifth instars have dark orange heads, first antennae segment was orange and next segments were darker. Thorax and abdomen were dark orange. Legs were pale orange with grey stripes. Scuttelar spine were dark orange and wing buds were brown and covers two third of its abdomen. Fifth instar nymphs will next turn into imagoes (Figure 2G & 2H).

Differences between male and female imagoes could be observed from their body size, abdomen shape, and pronotum color. Females were larger than males. Abdomen of males were smaller, flat, and abdomens were completely covered by wings at resting poses. Meanwhile, female abdomens were larger, round, and enlarged on sides causing abdomen to be unable to fully covered by wings at resting pose. New imagoes were pale orange and turn dark as imagoes age. Female imagoes have dark red pronotum while male imagoes have dark pronotum. Male imagoes turn color quicker than female imagoes (Melina *et al*., 2016b). Biological statistics of *H. bradyi* reared on cucumbers are shown in Table 1.

Life Table of *Helopeltis bradyi*

Life table contain basic information on insect population dynamic and can be used to determine potential reproduction at specific environments. Based on rearing results in this study, 304 eggs were produced from a imago pair of the second generation. Eggs were incubated for 6.33 ± 0.47 days (Table 1). There were 262 eggs of *H. bradyi* that hatched, 199 individuals reached second instar, 171 individuals reached third instar, 138 individuals reached fourth instar, 114 individuals reached fifth instar, and resulted in 53 males and 57 females. *H. bradyi* survivorship shows high mortality during early instar stages and gradually decrease as age increased. Insect populations are sensitive to changes of abioctic conditions, such as temperature. Change of parameters can affect growth and survivorship of insects (Schowalter, 2006). The first to fifth instar lasted for 2.13 ± 0.34 days; 2.07 ± 0.25 days; 2.13 ± 0.34 days; 2.33 ± 0.47 days; and 3.20 ± 0.40 days. *H. bradyi* nymph stage lasted for 11-13 days before turning into imagoes. Egg incubation and nymph development was shorter than the studies from Melina *et al.* (2016a) that was 8.50 days for first to fifth instar nymph 3.30 days; 2.20 days; 2.30 days; 2.90 days; and 4.30 days respectively. This can be caused due *H. bradyi* rearing did not use air conditioners. Lower or higher temperature compared to optimum temperatures may decrease development rate (Begon *et al.*, 2006). Temperature affect the duration of each instar and the number of individuals that go through each stages and reach imagoes (Du Plessis *et al.*, 2020).

Fast development rates and cause insect to reach reproduction stage sooner (Begon *et al.*, 2006). Development of each instar go through moulting. Lifespans of *H. bradyi* imagoes were counted since the day imago first emerged until they die. Male and female imagoes have different lifespans. Lifespan of male imagoes were 20-39 days with average of 32.33 ± 4.92 days; and females were 12-37 days with averages of 24.47 ± 6.71 days. Long lifespans may result in higher reproductions.

Life table (Table 2) shows life expectancy of *H. bradyi.* Highes mortality (dx) occured on eggs and first instar nymphs. Mortality proportions (qx) and life expectancy (ex) are used to forecase populations of an organism. If qx > ex, then populations will decrease; qx = ex then populations will be static; qx < ex populations will increase. In this study, qx < ex in all stages indicating that *H. bradyi* populations will increase. Stable age distribution (Lx) is the distribution estimated on populations stable birth and death patterns (Kakde *et al.,* 2014). Results from this study showed that stable age distribution of *H. bradyi* on eggs, 1–5 nymph instar, and imagoes were 283, 230.5, 185, 154.5, 126, 112, and 55 respectively.

Fecundity and Survivorship of *Helopeltis bradyi*

Average sex ratio was 0.93:1.08 for male:female. Sex ratio is essential in determining growth rate of a population (Srikumar & Bhat, 2012). More females cause insect populations to grow faster. Pre-oviposition duration of female *H. bradyi* imagoes was 2.2 ± 0.40 days after imagoes occured. Short pre-oviposition periods cause females to lay their eggs sooner. First eggs were laid by females at age of 19 days or 2 days after turning into imagoes. Offsprings of *H. bradyi* produced was 296 eggs during a females' lifetime and over a 22.9 ± 4.66 day oviposition period. Longer oviposition period will increase the number of eggs produced. Highest fecundity occurred age 22 and 23 with 16 egss.

Survivorship of *H. bradyi* can be shown in a survivorship curve (lx), while fecundity were represented as fecundity curve (mx), which both curve showed the respective variable across time (Figure 4). Surival proportion of *H. bradyi* was obtained from daily observation since nymph stages to imagoes. According to Price (1997), there are three types of survivorship curves that are type I, type II and type III. Type I shows higher mortality occurs during older stages, type II shows constant mortality, while type III shows higher mortality during early stages. Based on this study, highest mortality rate of *H. bradyi* occurred during the egg and nymph stages based on survivorship curve implying it to follow the type III curve. Survivorship curve showed that early pre-immature stages were susceptible to physical disturbances, such as cleaning and changing food. Newly hatched *H. bradyi* were fed with cucucmbers and requires adaptation to new food sources and living environment.

Demographic Statistics of *Helopeltis bradyi*

Demographic statistics are required to estimate population growth. Population growth were calculated based on survival potential (lx) and number of offspring for females (mx). Results showed that GRR of *H. bradyi* was 296 individuals from each generation. while Ro of *H. bradyi* was 196 individuals each female each generation. Population growth depends on net growth rate. Net growth rate is the number of female offspring produce by each female in each generation (Price, 1997). High Ro and GRR values show suitability of food source (Kurniawan, 2007). Intrinsic growth of constant environment and unlimited food source was shown by r value. Intrinsic growth (r) show constant population growth. The value r can be affected by environmental condition changes (Djanika *et al.*, 1998). The r value in this study was 0.18 individuals each female each day. This shows that populations are in unlimited conditions and can grow with accelarations of 18%. Values are affected by factors related to organisms life cycles, such as death, birth, and development time. Intrinsic growth can be used to predict long-term insect population growth (Hidayat *et al.*, 2019).

Average generation time (T) is the duration of an individual lifespan since eggs were laid to when females have laid half of their eggs. Average generation time of *H. bradyi* was 29.34 days. Smaller T value cause organisms to reproduce sooner. Doubling time of *H. bradyi* was 3.85 days. Low doubling time values increase gross reproduction rates (GRR) and net reproduction rates (Ro) during a certain time period. High *H. bradyi* fecundity affect the short time for a population to double its amount and increase intrinsic growth.

CONCLUSION

Life cycle of *H. bradyi* was 18.19 days. The duration of egg stage was 6.33 ± 0.47 days. Average length of nymph instar I was 2.13 ± 0.34 days, instar II 2.07 ± 0.25 days, instar III 2.13 ± 0.34 days, instar IV 2.33 ± 0.47 days, and instar V 3.20 ± 0.40 days. The lifespan of male imagoes were 32.33 ± 4.92 days, while for female imagoes was 24.60 ± 6.64 days. Preoviposition, oviposition stage, and post-ovipostion ages were respectively 2.2 ± 0.40; 22.9 ± 4.66; and 2.55 ± 0.80 days. Fecundity of *H. bradyi* was 296 each female. Survival curve of *H. bradyi* was considered as a type III that showed high mortality during earlier life stages. Demographic statistics of *H. bradyi* reared on cucumbers were as the following, Gross reproduction rate (GRR) as much as 296 individual per generation, net reproduction rate (Ro) was 196 individuals each female each generation, intrinsic growth (r) was 0.18 individuals each female each day, average generation length (T) as much as 29.34 days, and doubling time (DT) was 3.85 days.

Helopeltis bradyi was reared in laboratory conditions on cucumbers had potential to cause damage. Based on it demography, *H. bradyi* had the ability to reproduce and adapt well with relatively short doubling time that may result in many offspring. Therefore, should be monitored closely.

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Data shown in this study has not been published anywhere although this topic was part of the first author's Master thesis.

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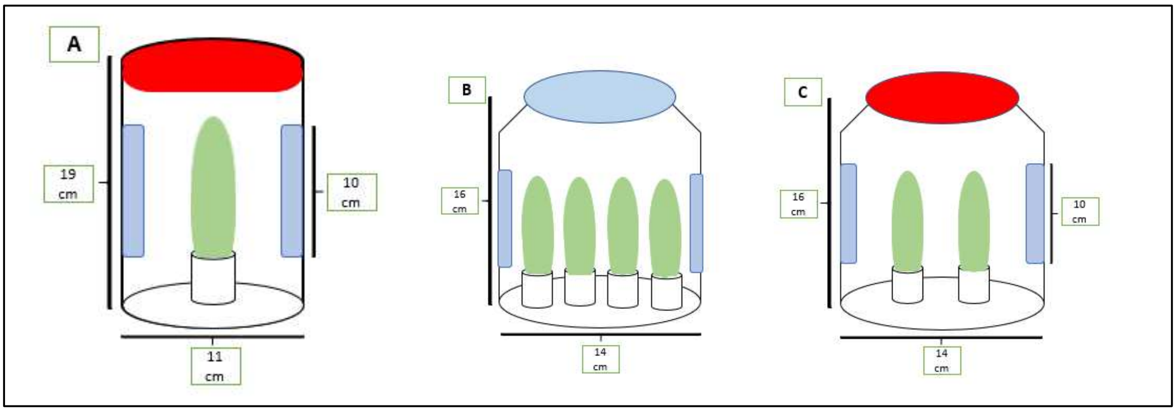

Figure 1. *Helopeltis bradyi* rearing container: imago container (A), egg incubation container (B), nymph containers (C)

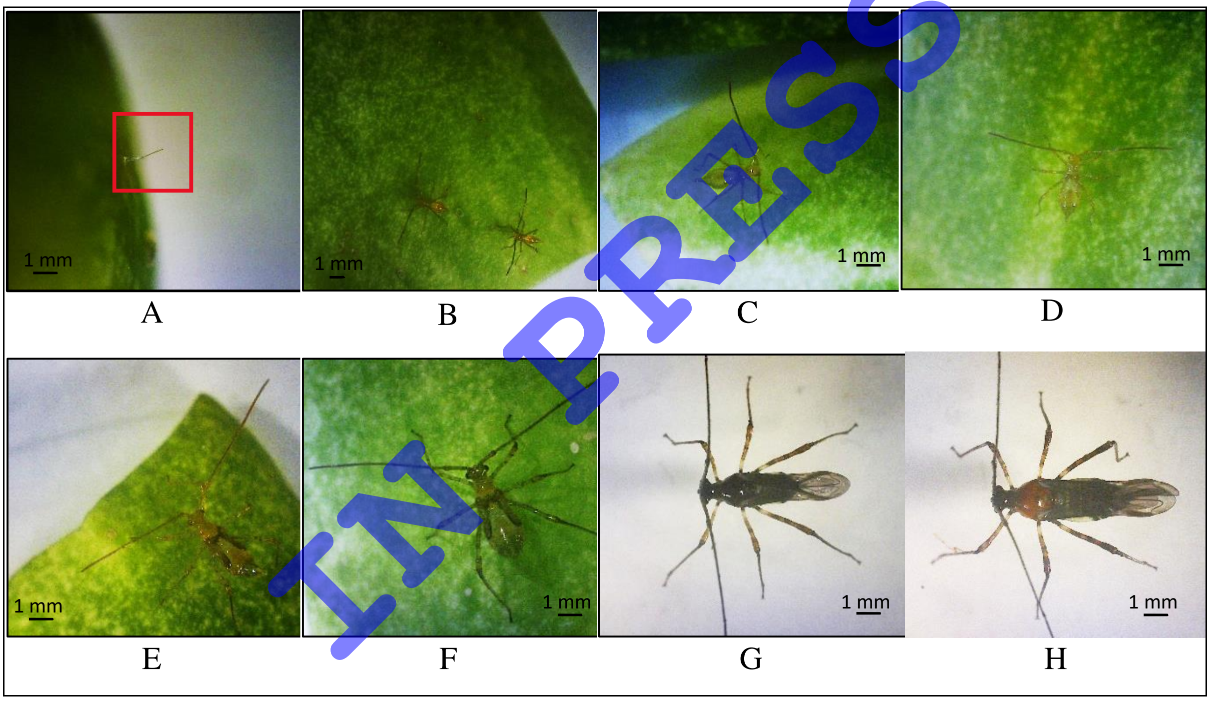

Figure 2. *Helopeltis bradyi* development stages on cucumber. Eggs in cucumber tissue (A), nymph instar I–V (B–F), male imago (G), and female imago (H)

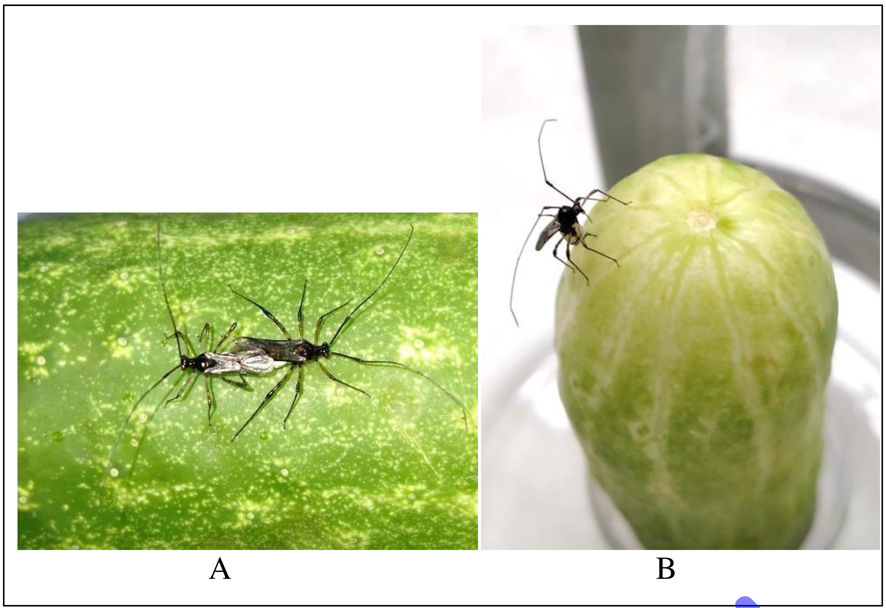

Figure 3. *Helopeltis bradyi* imago: copulation (A), oviposition (B)

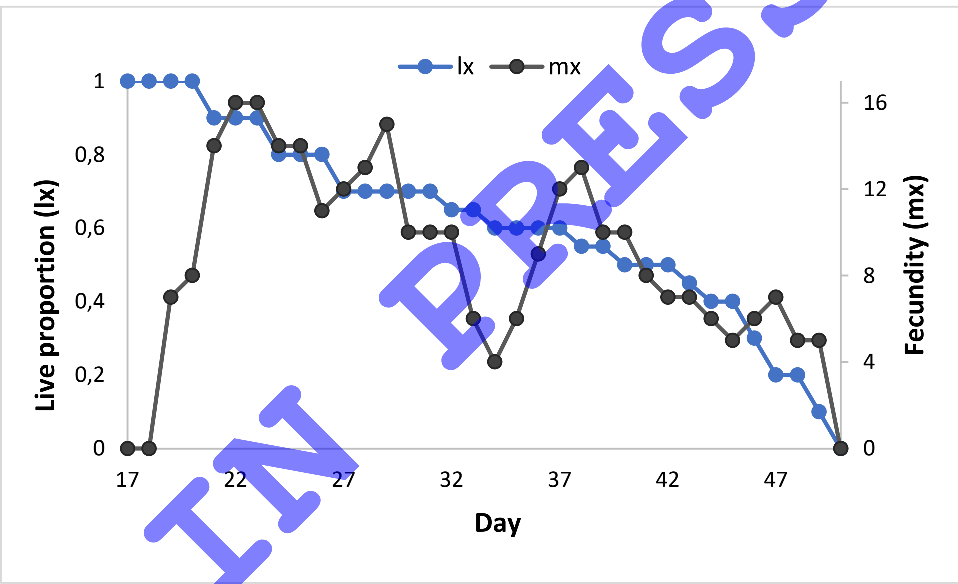

Figure 4. Survivorship curve of *Helopeltis bradyi*

Table 1. Biological statistics of *Helopeltis bradyi* reared on cucumbers

Average age (day)	Phase	Individuals (n)	Mean± SD (day)
0 – 6	Egg	304	6.33 ± 0.47
6 – 8	Instar 1	262	2.13 ± 0.34
8 – 10	Instar 2	199	2.07 ± 0.25
10 – 12	Instar 3	171	2.13 ± 0.34
12 – 14	Instar 4	138	2.33 ± 0.47
14 – 17	Instar 5	114	3.20 ± 0.40
17 – 49	Male	53	32.33 ± 4.92
17 – 42	Female	57	24.60 ± 6.64
	Pre-oviposisi	57	2.2 ± 0.40
	Oviposisi	57	22.9 ± 4.66
	Post-oviposisi	57	2.55 ± 0.80
	Fecundity (individuals)	296	

Table 2. Life tables of *Helopeltis bradyi* reared on cucumbers in laboratory

x	ax	dx	Lx	qx	Tx	ex
Egg	304	42	283	0.14	1146	3.77
Instar 1	262	58	230.5	0.22	863	3.29
Instar 2	199	28	185	0.14	632.5	3.18
Instar 3	171	33	154.5	0.19	447.5	2.62
Instar 4	138	24	126	0.17	293	2.12
Instar 5	114	4	112	0.04	167	1.46
Imago	110		55			

Notes: x: development stage, ax: living individulas, dx: dead individuals. Lx: stable age distribution, qx: mortality proportion, Tx: total individuals at age x and beside x, ex: life expectacy.

Table 3.Demographic parameter of *Helopeltis bradyi*

Parameter	**Value**	**Unit**
GRR	296	Individual/generation
Ro	196	Individual/female/generation
r	0,18	Individual/female/day
T	29,34	Day
DT	3,85	Day

Notes: GRR: Gross reproduction rate; Ro: net reproduction rate; r: instrinsic growth; T: average generation length; DT: doubling time.