

BIOLOGY AND LARVAL INSTAR DETERMINATION OF BLACKHEADED FIREWORM, *RHOPOBOTA NAEVANA* (HUBNER) (LEPIDOPTERA: TORTRICIDAE)

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KEYWORDS

Apple
Biology
Kashmir
Rhopobota naevana

Received on :

16.09.2015

Accepted on :

16.04.2017

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ABSTRACT

The studies on biology and number of larval instars of *Rhopobota naevana* (Hubner) in temperate region of Jammu and Kashmir were conducted from 2010-2012. The incubation period ranged from 3.78-4.59 days in different generations. Total larval development period ranged from 18.94-22.17 days in different generations, while as pupal period on an average lasted for 8.41 ± 1.21 days. Male and female showed an average longevity of 5.41 ± 0.73 and 7.22 ± 0.80 days respectively and fecundity per female ranged from 66.12-92.50 eggs in different generations. Thus whole life cycle completed in 38.29 - 47.80 days in different generations. Results also emphasised that development took place through holometamorphosis and close proximity between the mean, observed and calculated widths of head capsule and increase in widths of head capsules of different larval instars categorised them into five well defined instars. The mean geometric growth ratio of 1.54 using the head capsule width in successive larval instars as a parameter was obtained and it conformed well to Dyar's law

INTRODUCTION

Apple is the most dominant fruit crop of Jammu & Kashmir and accounts for 51% of total area of 2.72 lakh hectare under all temperate fruits grown in this state. Average productivity of commercially important apple cultivars is the highest in the country ranging from 10-12 tonnes/ha, but it compares poorly to the productivity of 20-30 tonnes/ha in horticulturally advanced countries of the world (SKUAST, 2013). One of the main causes of low productivity is ineffective control of insect pests. Majority of Insect pests that attack apple fruit belong to orders Hemiptera, Coleoptera, Lepidoptera and Diptera (Chadha, 1995). Among Lepidoptera extensive fauna of tortricid moths attack apple crop (Cross, 1996). Meszaros, et al. (1984) recorded 88 species of tortricid moths in the apple orchards of Hungary and 25 species have been recorded in apple orchards throughout the United Kingdom (Alford, 1984). In temperate region of Jammu and Kashmir, 64 species of tortricid moths have been reported (Razowski 2006), out of which only two are yet reported to attack apple crop. These are Codling moth, *Cydia pomonella* (Linnaeus), which occurs as severe problem in Ladakh region (Wadhi and Sethi, 1975) and Archips moth, *Archips pomivora* Meyrick which appears sporadically in Kashmir valley (Bhagat et al., 1994), while as Blackheaded fireworm, *Rhopobota naevana* (Hubner) which is first time reported to damage apple trees is a problem in young and unmanaged apple orchards of Kashmir valley. It overwinters on current season twigs as eggs that hatch in early spring. Larvae feed primarily on terminal foliage, webbing the terminals together, frequently destroying buds, and

skeletonising leaves often so severely that areas of the apple orchards turn brown and appear burned top of trees. They wriggle vigorously when disturbed and use silk to construct protected feeding sites by webbing together terminal leaves. For successful management it is necessary to predict the age of individuals and to determine the time taken by insect to develop from one life stage to another. Biology of some tortricid moths have been studied by various authors throughout the world, however reports on the biology and larval instar determination of Blackheaded fireworm, *Rhopobota naevana* (Hubner) are not available except some preliminary studies undertaken by Alford (1984), Cockfield and Mahr (1992), Fitzpatrick and Troubridge (1993), Cockfield et al. (1994), Maurice et al. (2000) and Sylvia and Averil (2005). Hence the present investigation was undertaken with the objective to study the biology, larval instar determination, morphometrics and life cycle of Blackheaded fireworm, *Rhopobota naevana* (Hubner) on apple in temperate region of Jammu & Kashmir during 2010-2012.

MATERIALS AND METHODS

The present investigation was undertaken at Division of Entomology, SKUAST-Kashmir Shalimar, during 2010-2012. Blackheaded fireworm, *Rhopobota naevana*, (Hubner) larvae were initially collected from the field brought to laboratory and reared at an ambient temperature (25 ± 5 °C) and relative humidity (70 ± 5 %). These larvae were kept in glass jars (20 x 15 cm) covered with muslin cloth and terminal shoots of apple

having cut ends immersed in 250 ml conical flask containing water to maintain the turgidity were provided them as food so as to obtain adults for further investigation. After pupation male and female pupae were identified taxonomically (Gadhiya *et al.*, 2014), thereafter isolated in pairs and confined to separate glass jars (20 x 15 cm). After adult emergence these were provided with 10 per cent sugar/ honey solution soaked in cotton swabs and tender shoots with their petioles wrapped with water soaked cotton were provided as ovipositional substrate. Observations on number of eggs laid by a female were recorded and freshly laid eggs were transferred along with a portion of leaf to a piece of filter paper seated over moist cotton in a Petri dish to study incubation period and hatching percentage. Newly hatched larvae were individually transferred to a piece of apple leaf placed on filter paper seated over moist cotton in a Petri dish and fresh leaves were provided them as food daily until pupation. In this way observations on larval duration, pre pupal period, pupal period, preoviposition period, oviposition period, post oviposition period, fecundity, viability of eggs, male and female adult longevity and female adult longevity were recorded (Maurice *et al.*, 2000; Mahr, 2005; Sylvia and Averill, 2005). Since larva roll the leaves and feed inside, it is difficult to find the cast head capsule (Kumar *et al.*, 2013), so Dyar's law (Dyar, 1890) was used to find out the moulting. For this purpose it was subjected to laboratory investigation on its development, life history and geometric growth ratio of successive instars. The development from egg to adult was described and their respective sizes were measured. This was done under a dissecting Stereo zoom Olympus microscope containing ocular grids calibrated with a stage micrometer. The mean width of head capsule in all larval instars were measured which yielded a mean consecutive geometric progression. The calculated or theoretical head capsule width was thus computed from the average mean geometric progression. The difference or the discrepancy between the observed and calculated or theoretical head capsule width was computed from pooled Chi-square value (χ^2) by using the Pearson's Chi-square test.

RESULTS AND DISCUSSION

Biological studies on developmental stages of egg, larval instars, pupae, adult (both male and female), pre-oviposition, oviposition, fecundity, post-oviposition period of *Rhopobota naevana* (Hubner) on apple are shown in Table 1. The present study revealed that after matting female on an average laid

78.94 ± 13.20 whitish disc shaped eggs in different generations. These results are in confirmation with the findings of Fitzpatrick and Troubridge (1993) who recorded the female fecundity of 70-80 eggs during first and second generations. The eggs were deposited singly or in small groups on terminal unopened leaves and young shoots. The mean incubation period under laboratory conditions was found as 4.18 ± 0.40 days while as mean fertility was observed as 74.76 ± 9.62 eggs. The present results are in close agreement with the observations made by Cockfield *et al.* (1994) who also observed incubation period of 6-15 days.

The larvae underwent five instars and a pupal stage to reach adulthood. The newly hatched larvae are greenish coloured with black head capsule, second instar larvae are greenish-yellow in colour, third instar larvae are greenish-grey in colour, with distinct spiracles and body segments, fourth instar larvae are light-grey in colour and other morphological characters are same as third instar and fifth instar full grown larva is greyish coloured with more prominent spiracles and abdominal segments. The mean duration of 1st, 2nd, 3rd, 4th and 5th instar larvae were 2.29 ± 0.07, 3.47 ± 0.17, 4.27 ± 0.38 and 4.78 ± 0.41 and 6.18 ± 0.79 days, respectively. The present findings are in accordance with the findings of Cockfield and Mahr (1992), who recorded five instars with same colour features on the body and also duration of instars. Total larval development period ranged from 18.94-22.17 days in different generations with an average of 21.01 ± 1.80 days. The present results are in close agreement with the observations made by Sylvia and Averill (2005) who reported larval development period ranged from 2.5-5.0 weeks. The duration of prepupal period ranged from 2.80-3.66 days in different generations with an average of 3.27 ± 0.43 days. Pupation took place in webbed leaves inside the thin silken cocoon and its duration ranged from 7.21-9.63 days in different generations with an average of 8.41 ± 1.21 days. Present observations are in agreement with Fitzpatrick and Troubridge (1993) which reported duration of first and second generation pupae as 10 and 13 days, respectively.

The male moth is small in size. Fore wings are subfalcate, with a distinct notch below apex, ground colour white to ochreous-white, suffused or overlaid with plumbeous and sparsely strigulated with brown or fuscous, markings brown to blackish brown, basal and sub-basal fasciae more or less confluent and forming a moderately well-defined basal patch. The outer edge is obliquely curved from costa to dorsum to form costal



A. Culture maintained in laboratory



B. Culture inside cage



C. Pupal cases in leaves



D. Pupae obtained from culture

Plate 1:

Table 1: Biological attributes of Blackheaded fireworm, *Rhopobota naevana* (Hubner)

S. No.	Developmental stage	Duration (Days)		
		Range	Mean \pm SD	
1	Pre-oviposition period	2.00 - 2.45	2.26 \pm 0.23	
2	Oviposition period	3.25 - 3.55	3.37 \pm 0.15	
3	Post-oviposition period	1.12 - 1.97	1.58 \pm 0.42	
4	Fecundity	66.12 - 92.50	78.94 \pm 13.20	
5	Incubation period	3.78 - 4.59	4.18 \pm 0.40	
6	Hatching (%)	64.16 - 82.96	74.76 \pm 9.62	
7	1 st instar larvae	2.21 - 2.34	2.29 \pm 0.07	
8	2 nd instar larvae	3.29 - 3.64	3.47 \pm 0.17	
9	3 rd instar larvae	3.83 - 4.53	4.27 \pm 0.38	
10	4 th instar larvae	4.35 - 5.18	4.78 \pm 0.41	
11	5 th instar larvae	5.26 - 6.66	6.18 \pm 0.79	
12	Total larval period	18.94 - 22.17	21.01 \pm 1.80	
13	Prepupal period	2.80 - 3.66	3.27 \pm 0.43	
14	Pupal period	7.21 - 9.63	8.41 \pm 1.21	
15	Mean adult emergence:			
	a. Male	5.42 - 5.87	5.65 \pm 0.22	
	b. Female	4.12 - 4.57	4.34 \pm 0.22	
	c. Sex ratio	1.18 - 1.42	1.30 \pm 0.12	
16	Adult longevity:			
	a. Male	4.62 - 6.07	5.41 \pm 0.73	
	b. Female	6.37 - 7.97	7.22 \pm 0.80	
17	Total life cycle:			
	a. Male	38.29 - 45.90	42.31 \pm 3.82	
	b. Female	40.06 - 47.80	44.11 \pm 3.88	

Table 2 : Body sizes of egg, different larval instars, pupa and adult of Blackheaded fireworm, *Rhopobota naevana* (Hubner).

Stage of Development	Number of specimens (n)	Length (mm)		Width (mm)	
		Range	Mean \pm SD	Range	Mean \pm SD
Egg	20	0.79 - 0.90	0.84 \pm 0.05	-	-
Larva: 1 st Instar	15	2.04 - 2.64	2.39 \pm 0.31	0.84 - 1.08	0.97 \pm 0.12
2 nd Instar	15	3.84 - 5.04	4.42 \pm 0.60	1.20 - 1.44	1.33 \pm 0.12
3 rd Instar	15	5.52 - 6.72	6.08 \pm 0.60	1.68 - 2.16	1.94 \pm 0.24
4 th Instar	12	7.20 - 8.40	7.90 \pm 0.62	2.16 - 2.40	2.25 \pm 0.12
5 th Instar	12	8.88 - 9.60	9.16 \pm 0.38	2.52 - 2.76	2.64 \pm 0.12
Pupa: Pre pupa	15	6.96 - 8.16	7.44 \pm 0.63	2.64 - 3.00	2.78 \pm 0.19
Male pupa	12	5.40 - 6.60	6.00 \pm 0.60	2.88 - 3.12	3.00 \pm 0.12
Female pupa	12	6.00 - 6.96	6.48 \pm 0.48	3.12 - 3.72	3.43 \pm 0.30
Adult: Male	10	11.28 - 11.88	11.52 \pm 0.31 (w.sp.)	3.96 - 4.32	4.12 \pm 0.18
Female	10	12.00 - 12.60	12.23 \pm 0.32 (w.sp.)	4.44 - 4.80	4.59 \pm 0.18

Table 3: Geometric growth ratio of Blackheaded fireworm, *Rhopobota naevana* (Hubner) larvae using head capsule width as a parameter.

Stage of Development	Number of Specimens (n)	Mean observed head capsule width (mm)	Geometric progression	Calculated head capsule width (mm)	χ^2
1 st Instar	10	0.28	0.42/0.28 = 1.50	0.28	
2 nd Instar	12	0.42	0.65/0.42 = 1.54	0.28x1.54 = 0.43	
3 rd Instar	14	0.65	1.02/0.65 = 1.56	0.43x1.54 = 0.66	0.0011
4 th Instar	15	1.02	1.60/1.02 = 1.56	0.66x1.54 = 1.01	
5 th Instar	15	1.6	2.45/1.60 = 1.53	1.01x1.54 = 1.55	
Adult	12	2.45		1.55x1.54 = 2.38	
Total	78	6.42	Mean G. ratio = 1.54	6.31	P > 0.01

χ^2 = Pearson's Chi-square test (P = 1)

fold. Hind wings are grey or light fuscous, darker apically, upper side with a patch of violaceous-grey scales on cell area and a similar conspicuous patch of coarse grey black scales on underside. Female moth is larger in size than male, other characters are similar to male but lacking the patches of specialized scales on the hind wing and costal fold on fore wing. Present findings are in agreement with the reports of

Bradley *et al.* (1979) who also reported the same external characters of adult moth.

Mating was noticed on the host plant between dusk and evening and was not observed day time. Maurice *et al.* (2000) also reported mating in late afternoon and evening. The preoviposition period ranged from 2-2.45 days with an average of 2.26 ± 0.23 days. Female started fertile egg laying one day



A. Female and male pupae

B. Rearing inside cage

C. Adult emergence inside cage

D. Mating inside cage

Plate 2:



A. Freshly laid eggs

B. Ready to hatch eggs

C. First instar larvae

D. Second instar larvae

E. Third instar larvae

Plate 3:

after mating and oviposition period lasted for 3.25-3.55 days with an average of 3.37 ± 0.15 days, while as post-oviposition period ranged from 1.12-1.97 with an average of 1.58 ± 0.42 days. Fitzpatrick and Troubridge (1993) also reported that females laid 75% eggs on the day after mating. Fecundity ranged from 66.12-92.50 eggs per female during different generations with an average of 78.94 ± 13.20 eggs. Male adult longevity ranged from 4.62-6.07 days with an average of 5.41 ± 0.73 days while as the female adult longevity ranged from 6.37-7.97 days with an average of 7.22 ± 0.80 days. Thus male was found to complete its life cycle in an average of 42.31 ± 3.82 days while as female in 44.11 ± 3.88 days. These findings are in agreement with those of Bradely *et al.* (1979), Alford (1984) and Sylvia and Averill (2005). Finally the study confirmed that *Rhopobota naevana* completes its life cycle in 38.29 - 47.80 days in different generations and overwinters in egg stage that hatches around late April to mid May. The first generation larvae generally occur between late May to early June, adults emerge and lay eggs from late June to early July, second generation larvae occur from mid July to early August and adult fly from early to mid August and third generation eggs are laid in mid August, larvae occur from later August to

early September, adult fly from mid to late September and lay eggs which overwinter, thus completes three generations per year. These findings are in agreement with Sylvia and Averill (2005), which reported three generations per year during warm years in North America.

The morphometric measurements on egg, larval instars, pupae and adults (both male and female) of *R. naevana* on Apple are shown in Table 2. In this study, the diameter of an egg varied from 0.79 to 0.90 mm with an average of 0.84 ± 0.05 mm, similar results were also reported by Sylvia and Averill (2005). The mean length and width of 1st, 2nd, 3rd, 4th and 5th instar larvae were 2.39 ± 0.31 mm and 0.97 ± 0.12 mm, 4.42 ± 0.60 mm and 1.33 ± 0.12 mm, 6.08 ± 0.60 mm and 1.94 ± 0.24 mm, 7.90 ± 0.62 mm and 2.25 ± 0.12 mm and 9.16 ± 0.38 mm and 2.64 ± 0.12 mm respectively. Present observations are in close agreement to those of Sylvia and Averill (2005), which reported the length of fully grown larvae as 7-9 mm. Prepupa on an average measured 7.44 ± 0.63 mm in length and 2.78 ± 0.19 mm in width while as the mean length and width of male and female pupae were observed as 6.00 ± 0.60 mm and 3.00 ± 0.12 mm and 6.48 ± 0.48 mm and 3.43 ± 0.30 mm respectively. The present findings are in agreement with



A. Fourth instar larvae

B. Fifth instar larvae



C. Pupal case

D. Female and male pupae

Plate 4:

the findings of Sylvia and Averill (2005) who also reported the mean length pupa as 6-7 mm. The male moth on an average measured 11.52 ± 0.31 mm across the expanded wings while as the mean wing span of female moth was found 12.23 ± 0.32 mm.

Dyar's law was used to find out the number of larval instars based on the head capsule width measurement are shown in Table 3. The results in this present study revealed that mean widths of head capsule in 1st to 5th larval instars were 0.28, 0.42, 0.65, 1.02 and 1.60 mm, respectively, which yielded a consecutive geometric progression during each stage ranging from 1.50-1.56 with an average of 1.54. The calculated or theoretical head capsule width was thus computed from the average mean geometric progression of 1.54. The calculated or theoretical head width although was not completely identical to observed head width but these were sufficiently close to each other. Slight variation might be due to less number of measurements of each instar. Using the Chi-square test, the computed pooled Chi-square value (χ^2) was 0.0011 with p-value > 0.01 . Therefore, the difference or the discrepancy between the observed and calculated or theoretical head capsule widths was not significant and the geometric growth ratio of 1.54 confirmed to Dyar's law. No earlier reports were found on the growth ratio of *Rhopobota naevana* elsewhere to determine its development.



A. Adult moth



B. Male and female moths resting position

C. Damage

Plate 5:

Finally it could be concluded that Blackheaded fireworm, *Rhopobota naevana* (Hubner) completes its larval stage through five distinct instars, undergoes hibernation in its egg stage and has complete three generations in temperate region of Jammu & Kashmir from May to October.

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