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Biology of pulse beetle, Callosobruchus chinensis (Linnaeus) on cowpea variety DC-15

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Abstract

Pulses are the second most important group of crops all over the world next to cereals. Among these, cowpea (*Vigna unguiculata* (L.) Walp.) assumes greater importance which is grown as vegetable, fodder and also used for seed purpose. The full yield potential of cowpea is hardly ever realized due to the attack of several pests, of which bruchid infestation takes the lion share of the total seed loss after harvest. Hence, the study of biology of *Callosobruchus chinensis* (Linnaeus) was undertaken during August-September 2017. The incubation period of the eggs under laboratory conditions ranged from 4 to 6 days with a mean of 4.6 ± 0.70 days. The total development period occupied 26 to 40 days with a mean of 30.90 ± 4.28 days. The females lived for a period of 8-12 days with a mean of 9.50 ± 1.58 days whereas the males lived for 7 to 11 days with a mean of 8.30 ± 1.25 days.

Keywords: Pulse beetle, cowpea, Callosobruchus chinensis, biology

1. Introduction

Among the different groups of crops grown across the world, pulses are the second most important next to cereals. They are very good sources of human dietary proteins (25-40%) and are much cheaper as compared to the non-vegetarian sources of protein like meat, egg and fish. Among these, cowpea (*Vigna unguiculata* (L.) Walp.) assumes greater importance which is grown as vegetable, fodder and also used for seed purpose. The choice of cowpea as vegetable can be owed to its high palatability, nutrient content and negligible concentration of toxic metabolites. It is rich in proteins (22-24%), iron (0.005%), calcium (0.08-0.11%) and essential amino acids.

The full yield potential of cowpea is hardly ever realized due to the attack of several pests, of which bruchid infestation takes the lion share of the total seed loss after harvest. Bruchids damage the seeds quantitatively by causing grain losses by consumption and qualitatively by contaminating the grains with excrement and fragments. The infested seeds are riddled with circular holes leading to loss of quality and viability. Development of a single larva in a pulse grain can lead to 8 to 22 percent weight loss and they are capable of destroying all cowpea grains by causing upto 100 percent losses within few months of storage ^[1].

Besides the direct damage inflicted to the stored legume grains, bruchid infestation also creates conditions that invites secondary infestation by rot organisms mainly fungi and subsequent mycotoxin contamination. Infested seeds are rendered unfit for human consumption as well as for sowing purposes due to quality and germination losses and mould growth ^[2]. Increased damage to seeds and deposition of metabolic wastes such as uric acid produced by bruchids also lead to reduction in germination of pulse seeds. The work on biology, varietal screening and management of pulse beetles in storage has been reviewed by Balikai and Neenu Augustine (2018) ^[3]. Keeping these points in view, a laboratory study was undertaken during August-September, 2017 to trace the life stages of *Callosobruchus chinensis* (Linnaeus) on cowpea in storage.

2. Materials and Methods

For initiation of culture, the adults of pulse beetles were collected from infested seeds of cowpea from the University storehouses. The stock culture was maintained on cowpea by releasing 10 pairs of freshly emerged beetles separately in plastic jars whose mouths were covered with muslin cloth and fastened by rubber band. Fresh grains were provided periodically for the development of beetles. The adults of *C. chinensis* was identified by using the taxonomic key ^[4].

The pure culture was raised by infesting insect-free cowpea seeds with freshly emerged single mating pair of beetles in plastic jars. The culture was maintained in the laboratory throughout the experimental period.

The biology of *C. chinensis* under laboratory conditions was studied on the cowpea variety DC-15 with respect various parameters like pre-oviposition and oviposition periods, incubation period, grub period, pre-pupal and pupal periods, developmental period, fecundity and adult longevity for male and female. The length and breadth of ten eggs, grubs of all instars, pre-pupa, pupa and adult male and females each were measured by using a mono-ocular microscope with the help of ocular and stage micrometers. The range and average for all the above parameters were worked out to draw the inference.

3. Results

3.1 Pre-oviposition period

Pre-oviposition period, *i.e.*, the time period between first mating and deposition of first egg was observed to spread over 4 to 9 hours with an average of 6.10 ± 1.66 hours (Table 1).

Table 1: Duration of different developmental stages of
Callosobruchus chinensis on cowpea during August-September,
2017

Sl.	Life Stages	Duration (days)		
No.		Range	Mean ± SD (n=10)	
1	Pre-oviposition period in hours	4-9	6.10 ± 1.66	
2	Oviposition period in days	5-10	7.10 ± 1.66	
3	Fecundity in numbers	75-100	86.50 ± 7.95	
4	Incubation period in days $4-6$ $4.60 \pm$		4.60 ± 0.70	
5	Grub period in days			
	First instar grub	3-5	3.50 ± 0.71	
	Second instar grub	3-6	4.40 ± 1.17	
	Third instar grub	4-6	4.80 ± 0.92	
	Fourth instar grub	4-7	5.50 ± 1.08	
	Total grub period	14-23	18.20 ± 3.58	
6	Pre-pupal period in days	2-3	2.60 ± 0.52	
7	Pupation period in days	6-8	6.80 ± 0.63	
8	Total developmental period in days	26-40	30.90 ± 4.28	
9	Adult longevity in days			
	Male	7-11	8.30 ± 1.25	
	Female	8-12	9.50 ± 1.58	

SD- Standard deviation, n = Sample size

3.2 Oviposition period and fecundity

In the present study, as many as nine eggs per cowpea seed was observed with more number of eggs on bold seeds. The oviposition period lasted for 5 to 10 days with a mean of 7.10 \pm 1.66 days. The highest number of eggs were observed to be laid on the second day of oviposition period; thereafter the number of eggs laid declined gradually till the end of oviposition period. The total number of eggs laid per female of *C. chinensis* on cowpea seeds varied from 75 to 100 eggs with an average of 86.50 \pm 7.95 eggs (Table 1).

3.3 Incubation period

The incubation period of the eggs of *C. chinensis* on cowpea under laboratory conditions ranged from 4 to 6 with a mean of 4.6 ± 0.70 days (Table 1). The eggs were laid singly which were oval, white and smooth in appearance. The length of the egg ranged from 0.48 to 0.58 mm with an average of 0.52 mm. It was 0.30 to 0.34 mm broad with an average of 0.32 mm (Table 2). The freshly laid eggs appeared transparent and were glued to the seed surface by a secretion from the mother beetle. The hatching of eggs were determined by the change in colour of the eggs. The hatched eggs turned to creamish white colour due to the accumulation of frass inside the egg.

 Table 2: Morphometric data of Callosobruchus chinensis on cowpea in storage

	Lengt	th (mm)	Breadth (mm)				
Life stages	Range	$\begin{array}{c} Mean \pm S.D \\ (n=10) \end{array}$	Range	Mean ± S.D (n=10)			
Egg	0.48-0.58	0.52 ± 0.04	0.30-0.34	0.32 ± 0.01			
First instar grub	0.46-0.55	0.50 ± 0.03	0.28-0.33	0.30 ± 0.02			
Second instar grub	0.70-0.82	0.77 ± 0.04	0.53- 0.59	0.57 ± 0.02			
Third instar grub	1.02-1.35	1.15 ± 0.11	0.67-0.95	0.79 ± 009			
Fourth instar grub	2.00-3.80	2.62 ± 0.58	1.22-2.16	1.60 ± 0.34			
Pre-pupa	3.76-3.90	3.83 ± 0.07	1.86-2.06	1.97 ± 0.06			
Pupa	2.92-3.75	3.24 ± 0.31	1.76-2.90	1.95 ± 0.13			
Adult male	3.22-3.90	3.49 ± 0.25	1.60-1.96	1.81 ± 0.12			
Adult female	3.52-4.60	4.00 ± 0.42	1.60-2.09	2.01 ± 0.13			
SD- Standard deviation $n - Sample size$							

SD- Standard deviation, n = Sample size

3.4 Grub

During its development period, the grubs of *C. chinensis* moulted three times thus giving rise to four instars. The four instars were identified based on the size of grub and castings of head capsule.

The first instar grub was small, opaque and creamish yellow in appearance. It was 0.46 to 0.55 mm long with a mean of 0.50 mm and had a breadth of 0.28 to 0.33 mm with an average of 0.30 mm (Table 2). The brown coloured head and body annulations were clearly visible. It was also characterised by a pair of pro-thoracic plates and the thoracic legs were represented by conical stumps. It was construed from the tunnelling pattern that after emergence, the first instar larva bored the seed vertically for a short distance; immediately after the second moult it took a turn horizontally. The duration of first instar ranged from 3 to 5 days with mean of 3.50 ± 0.71 days (Table 1).

The second instar grub was similar to the first instar except for its size and absence of pro-thoracic plates. Regarding size, it had a length of 0.70 to 0.82 mm with an average of 0.77 mm and a breadth of 0.53 to 0.59 mm with an average of 0.57 mm (Table 2). It was also recognised by the presence of casting of head capsule of the first instar. The body was stout and curved with the thoracic region characteristically larger than the posterior end. The brownish head was not distinct and was represented by the presence of mouth parts. The larva followed a horizontal path of boring to avoid getting deeper into the seed. The second instar larval duration ranged from 3 to 6 days with average of 4.40 \pm 1.17 days (Table 1).

The third instar grub was recognized based on size and the presence of castings of head capsules of the first and second moults which were seen sandwiched by faecal pellets. The larval length varied from 1.02 to 1.35 mm with an average of 1.15 mm and it was 0.67 to 0.95 mm broad with a mean of 0.79 mm (Table 2). It took 4 to 6 days for its development with a mean of 4.80 ± 0.92 days (Table 1). The final stage of grub was 2.00 to 3.80 mm long with an average length of 2.62 mm and was 1.22 to 2.16 mm broad with an average of 1.60 mm breadth (Table 2). It was also distinguished on the basis of the presence of three castings of head capsules seen as sandwiched between the faecal pellets of the preceding and existing instars. The grub fed deeper into the seed extending upto the seed coat where it left a thin layer of testa appearing

like a circular window.

Towards the end of larval period, the fourth instar grub constructed a pupal chamber which was oval in shape and was prepared by compacting faecal matter against the walls of the tunnel. Finally, it stopped feeding and became inactive facing the circular window facilitating adult emergence. The duration of this instar was 4 to 7 days with an average of 5.50 \pm 1.08 days. The total larval period of *C. chinensis* occupied 14 to 23 days with a mean of 18.20 \pm 3.58 days on cowpea under laboratory conditions (Table 1).

3.5 Pre-pupal and pupal periods

Pre-pupa is an inactive stage that comes in between the grub and pupal stages. Unlike the larval stages, the body divisions of pre-pupa were distinct and the abdominal portion is markedly broader than the thorax region. The length of prepupa was observed to be 3.76 to 3.90 mm with an average of 3.83 mm and the breadth was observed to be 1.86 to 2.06 mm with a mean of 1.97 mm (Table 2). The pre-pupal duration was 2 to 3 days with a mean of 2.60 ± 0.52 days (Table 1).

The pre-pupa moulted to give rise to an exarate pupa which was cream coloured. The length ranged from 2.92 to 3.75 mm with an average of 3.24 mm and the breadth ranged from 1.76 to 2.90 mm with a mean of 1.95 mm (Table 2). The appendages were free but held close to the body. As it reached the end of pupal period, the colour was observed to change to brown. The pupal development ranged from 6 to 8 days with an average of 6.80 ± 0.63 days. Total development period, *i.e.*, the number of days from egg to adult emergence of *C. chinensis* occupied 26 to 40 days with a mean of 30.90 ± 4.28 days (Table 1).

3.6 Adult emergence and mating behaviour

The adults emerged through the characteristic circular window which was cut by the last instar grub before pupation. They came out by pushing the circular lid by the head which came out first. The adult males were 3.22 to 3.90 mm long with an average of 3.49 mm and the breadth ranged from 1.60 to 1.96 mm with a mean of 1.81 mm. Adult females were 3.52-4.60 mm long with a mean length of 4.00 mm and were 1.60 to 2.09 mm broad with an average breadth of 2.01 mm (Table 2). The male antennae were pectinate with 4-10 segments conspicuously expanded anterolaterally whereas the female antennae were serrate. The antennae of both sexes were with 4-11 segments and dark brown in colour. Pygidium of female was covered with white coloured setae.

After emergence, the males which were comparatively more active, chased the females for mating. The females expressed resistance to the mating attempts by pushing the males away with hind legs. After several attempts the males succeeded in grasping the females and established copulation. Repeated matings were observed in case of both the sexes.

3.7 Adult longevity

It was observed that the females of *C. chinensis* lived longer than the male beetles. The females lived for a period of 8 to 12 days with a mean of 9.50 ± 1.58 days whereas the males lived for 7 to 11 days with a mean of 8.30 ± 1.25 days.

4. Discussion

In the present study, pre-oviposition period for *C. chinensis* on cowpea was observed to spread over 4 to 9 hours with an average of 6.10 ± 1.66 hours which is in close agreement with Chakaraborty *et al.* (2015) ^[5] who recorded a pre-oviposition

period of 6.36 hours and Singh (2017) ^[6] reported 5.80 hours on green gram. The oviposition period lasted for 5 to 10 days with a mean of 7.10 \pm 1.66 days which is in confirmation with Siddaraju (1994) ^[7] who recorded 5-10 days with a mean of 7.62 days and Chakraborty *et al.* (2015) ^[5] who reported an oviposition period of 6.18 \pm 1.39 days. The fecundity of the test insect recorded in the present study varied from 75 to 100 eggs with an average of 86.50 \pm 7.95 eggs. These results are supported by the findings of Singh (2017) ^[6] and Singh *et al.* (2017) ^[8] who observed that the average number of eggs laid by females of *C. chinensis* were 90.0 and 89.7, respectively.

The incubation period of the eggs of *C. chinensis* on cowpea under laboratory conditions ranged from 4 to 6 days with a mean of 4.6 ± 0.070 days. These findings confirm with the observations of Singh *et al.* (2017) ^[8] who reported an average incubation period of 4.17 days. However, this contradicts the findings of Chakraborty *et al.* (2015) ^[5] who reported a longer incubation period of 7.03 \pm 0.54 days in the first generation and 7.35 \pm 0.44 days in the second generation. This might be due to change in geographical location, climate and host seeds.

The observations on larval instars indicated that the grubs moulted three times and the four instars were distinguished based on the size of larvae and castings of head capsule. The boring pattern of the grubs observed in the present investigation is comparable with those of Howe and Currie (1964)^[9] and Sibi (2003)^[10]. As such literature on the morphological and morphometric characters of grubs of C. chinensis is scanty. However, the findings are in confirmation with those of Sibi (2003) [10] and Vidyashree and Thirumalaraju (2015) ^[11] who reported similar observations in C. analis and C. maculatus. The pupal development ranged from 6 to 8 days with an average of 6.80 ± 0.63 days which is in close agreement with the observations of Ahmad et al. (2016)^[12] who reported an average pupal period of 6.51 days for C. chinensis on chickpea. The total development period of C. chinensis occupied 26 to 40 days with a mean of $30.90 \pm$ 4.28 days. This is in line with the studies of Mahoviya (2017) ^[13] who reported a developmental period of 29.5 days on peas and 29 days on kidney beans.

The adults emerged by pushing the circular window which was cut by the last instar grub before pupation. After emergence the males chased the females for mating; but they were observed to resist the mating attempts. The males succeeded in grasping the females after several attempts and established copulation. Similar observations regarding adult emergence and mating behaviour were recorded by Ramesh (1993) ^[14], Siddaraju (1994) ^[7] and Vidyashree and Thirumalaraju (2015) ^[11]. The females lived for a period of 8-12 days with a mean of 9.50 ± 1.58 days whereas the males lived shorter *i.e.*, 7 to 11 days with a mean of 8.30 ± 1.25 days. Observations regarding the longevity of males and females corroborate with the findings of Sibi (2003) ^[10] and Mandal and Konar (2006) ^[15] who reported that the females lived longer compared to females.

5. Conclusions

Cowpea is one among the most preferred hosts of *C. chinensis* in storage. The incubation period of the eggs under laboratory conditions ranged from 4 to 6 days with a mean of 4.6 ± 0.70 days. The grubs moulted three times thus giving rise to four instars which were distinguished based on the size of larvae and castings of head capsule. The total developmental period occupied 26 to 40 days with a mean of 30.90 ± 4.28 days. The

females lived for a period of 8-12 days with a mean of 9.50 \pm 1.58 days whereas the males lived for 7 to 11 days with a mean of 8.30 \pm 1.25 days.

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