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# Generation- wise Comparative Biology of Pulse Beetle, *Callosobruchus maculatus* on different Stored Pulses

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**ABSTRACT: Pulse beetle, *Callosobruchus maculatus* is a major storage pest of various pulses. The studies on biology of *C. maculatus* on different pulses with emphasis on different generation provide more information for better management practices. So, in this study, generation-wise comparative biology of *C. maculatus* in different pulses such as cowpea, green gram, black gram, chickpea, soybean and pea were evaluated. The results revealed that, in a year, *C. maculatus* completed eight generations in cowpea, green gram and pea, seven generations on black gram and six generations in chickpea and soybean. The pre oviposition period, post oviposition period and incubation period of *C. maculatus* on six different pulses showed no significant difference which was in the range of 6.3 to 6.7 hours, 1.9 to 2.1 days and 3.1 to 3.3 days, respectively. The longest oviposition period, adult longevity of male and female was recorded ongreen gram which was 6.6 days, 8.8 days and 9.3 days, respectively while the shortest oviposition period, adult longevity of male and female was recorded on chickpea which was 5.0 days, 7.2 days and 7.6 days, respectively. The longest developmental period was recorded on chickpea (37.2 days) while the shortest developmental period was recorded on green gram (26.2 days). In addition to biological parameters, the ovipositional preference of pulse beetle was also studied. In the choice test, the maximum ovipositional preference of *C. maculatus* was recorded on green gram (19.25 % eggs) while the minimum ovipositional preference was recorded on chickpea (7.00 % eggs). In the no choice test also, the maximum ovipositional preference was recorded on green gram (87.00 % eggs) while minimum ovipositional preference was recorded on chickpea (74.00 % eggs).**

**Keywords:** Pulses, pulse Beetle**,** *Callosobruchus maculatus*, biology, oviposition, preference.

## INTRODUCTION

Pulses are a major protein supplement for human beings. They are not only an excellent source of protein (20-40 %), carbohydrates (50-60 %) and minerals (2-4 %) but also provide calories almost equal to that of cereals to the tune of 300-540 kcal /100 g (Ofuya and Akhidue 2005). The cultivation of pulses ensure global food security and it is threatened by several constraints (Murugesan *et al*., 2021). One of the most important constraints in pulses production is susceptibility of pulses to pests and diseases in both field and storage conditions. In storage, the important pest is pulse beetle, which comes under the family Bruchidae. Bruchids had co-evolved with different dried legumes in storage conditions (Rodriguez, 2018). Among the Bruchids, *C. maculatus* causes approximately 50 % losses during storage of cowpea and the losses due to *C. maculatus* in various pulses ranged from 30-40 % (Caswell, 1981). In India it is a serious pest of chickpea, green gram, moth bean, peas, cowpea, lentil, cotton, sorghum and maize seeds (Singh *et al*., 1989). The various kinds of losses due to *C. maculatus* infestation in pulses are quantitative and qualitative losses, losses of seed viability and damage to the storage containers (Bhargava and Kumawat 2010).

Generally, pulse beetle are more prolific, breed and increases their population within short span (Seni and Mishra 2022). Adult beetle don't feed on pulses and females of *C. maculatus* lay eggs on the surface of the pulses (Ahmad *et al*., 2018). On emergence, the larva enters inside the seeds and feed their inner contents. So, the development of the pulse beetle is completed inside the seeds. The development of pulse beetle mostly depend on the nutritive value of the legumes (Nwosu and Ikodie 2021). So, the female's decision to choose proper pulse is determined by the environment within which its progeny will complete the development. The ovipositional preference of pulse beetle is also dependent upon the resource quality as the females prefer high quality resources (Mitchell, 1975). Integrated Pest Management (IPM) involves understanding interactions between associated stored product environment, insects associated with stored products and replacing most of the chemical applications with cost effective nonchemical alternatives. To support IPM in stored pulses against *C. maculatus*, there is a need to understand the biology with respect to different generations as duration of pulses and food grains plays key role in storage. Though few studies are available with respect to biology of pulse beetle in various pulses (Nisar *et al*., 2021), still the study emphasis on generation- wise biology in different pulses is lacking. Chickpea, black gram, green gram, cowpea, pea and soybean are the major pulses in India and World (Mannava *et al*., 2022). So, our study focuses on the generation-wise biology of *C. maculatus* in these pulses in different generations. In addition to biology, ovipositional preference studies were also conducted for better understanding of host preference of pulse beetle.

## MATERIALS AND METHODS

**Culture of insect:** The inoculum of *C. maculatus* was obtained from the College of Agriculture, Central Agricultural University, Imphal. The rearing of insects and the stock culture was maintained in disinfested green gram seeds at the laboratory. The grain with egg was separated and it was kept in glass vials individually for adult emergence which was used to study the generation- wise comparative biology and ovipositional preference on different pulses.

**Separation of sex:** The male and female insects of *C. maculatus* were separated based on the morphological characteristics of adults of *C. maculatus* i.e. The female adult is dark brown in colour whereas the male adult is light brown colour (Khare, 1993). Also, the abdomen is shorter and the terminal segment is sharply curved downwards in males whereas the abdomen is comparatively longer and the terminal segment is only slightly bent downwards in females (Bandaara and Saxena 1995).

**Generation- wise comparative biology of *C. maculatus* in different pulses:** The comparative biology of *C.maculatus* was carried out under laboratory conditions on cowpea, green gram, black gram, chickpea, soybean and pea by using the method Zalavadia (1971) with slight modifications. This experiment was replicated five times in Completely Randomized Design (CRD). The same day emerged adult male and female was collected from glass vial and one pair of male and female was released into Petriplate which was having 20 seeds of cowpea, green gram, black gram, chickpea, soybean and pea. After Mating, the time has been taken for egg laying by female was recorded as pre oviposition period. The Petri plate with insects were replaced by new pulse grains for observing the number of eggs laid by an individual female till its oviposition period and such a way that the oviposition period and fecundity was recorded. The period after its oviposition to death of pulse beetle was recorded as post oviposition period.

Those seeds which contain eggs were collected and kept in separate Petri plate for further observations such as incubation period and developmental period (larval period + pupal period). 10 seeds were kept in each petriplate for such observations. The adults emerged from these seeds were used for observation of adult longevity and one pair from these were used for next generation by applying same procedure of first generation. The needed observations such as incubation period, developmental period, longevity of male and female, pre-oviposition, oviposition and post oviposition period and fecundity of *C. maculatus* were recorded in different generations from different pulses. Likewise, the lifecycle of Pulse beetle, *C. maculatus* in all generations from different pulses were studied and compared.

**Ovipositional preference of *C. maculatus* in different pulses:** The ovipositional preferential studies were done by following the method Soumia *et al.* (2017) with slight modifications. The free choice and no choice tests were conducted as follows.

**Free choice test:** Hundred grains from each pulse were kept in Petri plate separately. One pair of newly emerged adults were released in these Petri plates. Eggs laid after the release of insects were counted. This experiment was replicated four times in Completely Randomized Design.

**No choice test:** Ovipositional preference of *C. maculatus* was studied in Petri plate having the radius of 11 cm. Six equal compartments were made in the Petri plate by fixing strips of paper in a radial manner. Hundred grains from each of the pulses *viz.*, cowpea, chickpea, black gram, green gram, Pea and soybean were kept in these compartments in a randomized manner at equal distance from the centre. One pair of newly emerged adults of *C. maculatus* was released in the centre of the Petri plate. This experiment was replicated four times in Completely Randomized Design. The eggs laid after the release of insects were counted in each pulse.

**Data Analysis:** The data was analyzed with the help of one-way analysis of variance. When F values were found significant, then treatment means were compared by using critical difference values at 5% level of probability.

## RESULTS AND DISCUSSION

**Pre oviposition, oviposition and post oviposition period of *C. maculatus*:** In our study, the pulse beetle, *C. maculatus* completed eight generations in cowpea, green gram and pea, seven generations in black gram and six generations in chickpea and soybean. So, all the biological parameters were recorded for these generations. The pre and post oviposition period showed no significant difference whereas the oviposition period showed significant difference between the pulses. The mean pre oviposition period of *C. maculatus* from all the generations in six different pulses (Fig. 1) were in the range of 6.0 to 7.2 hours and were statistically non-significant. The mean period of pre oviposition was in the range of 6.3 to 6.7 hours which is in partial conformity with the findings of Zalavadia (1971) who reported that the average pre ovioposition period of *C. maculatus* was 5.10 hours from different pulses. The mean oviposition period of *C. maculatus* (Fig. 2) was longest on green gram (6.6 days) followed by pea (6.1 days) and cowpea (6.0 days) while the mean oviposition period of *C. maculatus* was shortest on chickpea (5.0 days) followed by soybean (5.2 days) and black gram (5.4 days). Zalavadia (1971) reported that the average oviposition period of *C. maculatus* was 4.5 days in different pulses which is almost similar to the results obtained from the first generation of our findings. The oviposition period of *C. chinensis* was 8.0 days on green gram and chickpea reported by Varma and Anandhi (2018); Kumar and Kumar (2018), respectively, collaborates with the present findings. The range of post oviposition period (Fig. 3) on six different pulses was recorded as 1.4 to 2.6 days and the mean post oviposition period was in the range of 1.9 to 2.1 days. Vyas (2004) reported that the post oviposition period of *C. chinensis* was 1.56± 0.58 days on cowpea which collaborates with present findings. Similarly, Kumar and Kumar (2018) observed that the post oviposition period of *C. chinensis* was 2.2 days on chickpea which also collaborates with present findings.

**Incubation and developmental period of *C. maculatus*:** The incubation period (Fig. 4) was also found to be non-significant in different pulses in different generations and the mean incubation period was in the range of 3.1 to 3.3 days. The previous findings show that the incubation period of *C. chinensis* was reported to be 5.0 days (Singh and Kumari 2000) and 4.0 days (Hosamani *et al.*, 2018) in different pulses. The developmental period (larval + pupal period), male and female adult longevity and fecundity of *C. maculatus* showed significant difference between the pulses in different generations. The mean developmental period of *C. maculatus* (Fig. 5) was longest in chickpea (37.2 days) followed by soybean (35.5 days) and black gram (30.1 days). Whereas shortest developmental period was recorded on green gram (26.2 days) followed by pea (27.2 days) and cowpea (27.8 days). Development and life table of *C. maculatus* vary with the pulses (Bidar *et al*.,2021) and our study also revealed similar results. The present findings are in conformity with the findings of Patel *et al*. (2005) who recorded the shortest developmental period of *C. maculatus* on green gram (33.51 days) followed by cowpea (34.52 days) and the longest developmental period of *C. maculatus* on chickpea (38.91 days) followed by pea (38.7 days). Also, the present findings are in partial conformity with the findings of Shivanna *et al*. (2011). Their reports show that the shortest mean developmental period of *C. maculatus* was found on cowpea (29.33 days) followed by green gram (29.67 days) while the longest period was found on black gram (32.67 days) followed by chickpea (31.33 days). Similarly, Swella and Mashobozy (2009) also reported the shortest developmental period of *C. maculatus* in cowpea (25.2 days) followed by garden pea (30.5 days) and green gram (31.4 days) while the longest period in soybean (34.6 days) followed by chickpea (34.0 days) and black gram (33.6 days).

**Adult longevity of *C. maculatus*:** The longest male adult longevity was recorded in green gram (8.8 days) followed by pea (8.3 days) and cowpea (8.2 days) while the shortest adult longevity of male of *C. maculatus* was recorded on chickpea (7.2 days) followed by soybean (7.4 days) and black gram (7.8 days) (Fig. 6). Male adult longevity of *C. chinensis* was reported to be 11 days on green gram (Varma and Anandhi, 2010) which is almost like 9.6 days on green gram obtained in the fourth generation in our findings. Neog (2012) also found that the longest male adult longevity of *C. chinensis* was recorded on green gram (10.00 days) and shortest male adult longevity was recorded on soybean (6.56 days) which collaborates with present findings.

Like male longevity, the longest female longevity (Fig. 7) was also recorded in green gram (9.3 days) followed by pea (8.8 days) and cowpea (8.7 days) while the shortest female longevity was recorded in chickpea (7.6 days) followed by soybean (7.7 days) and black gram (8.2 days). The female longevity of pulse beetle is more compared to male (Singh *et al*., 2021). Similar results were obtained in the present investigation. The female adult longevity of *C. maculatus* was reported to be 8-13 days on soybean (Sharma *et al.*, 2007) which is almost similar to 8.0 days on soybean obtained in the second, third and fourth generation in our findings.

**Fecundity of *C. maculatus*:** The fecundity of *C. maculatus* (Fig. 8) was maximum on green gram (86.0 eggs) followed by cowpea (83.4 eggs) and pea (83.1 eggs) while the minimum fecundity was recorded on chickpea (74.2 eggs) followed by soybean (76.2 eggs) and black gram (79.0 eggs). The present findings are in conformity with the findings of Kazemi *et al*. (2009) who reported that the maximum fecundity of *C. maculatus* was recorded on green gram (88.47 eggs) followed by cowpea (57.12 eggs) while the minimum fecundity was recorded on chickpea (39.13 eggs) followed by lentil (55.47 eggs). Srivastava and Subramanian (2016) reported that the fecundity of *C. maculatus* was in the range of 80- 100 eggs on different pulses, which also collaborates with our findings. The fecundity of *C. maculatus* was reported to be 59.5 eggs on soybean (Sharma *et al.,* 2007) which is almost similar to 62.8 eggs on soybean obtained in the first generation in our findings. Previous studies had documented the maximum fecundity of on cowpea (Dwivedi *et al.,* 2020), also corroborates our investigation.

**Ovipositional preference:** The percent egg laid by *C.maculatus* on different pulses in no choice and choice test are depicted in table 1. The results showed that there was a significant difference in oviposition of *C.maculatus* in different pulses. Green gram showed maximum ovipositional preference (19.25 % egg deposition) by *C.maculatus* in choice test followed by cowpea (16.50 % eggs). The cowpea was statistically at par with green gram. The percentage of egg laid on pea was 15.75 % which did not differ significantly from cowpea. The minimum ovipositional preference was observed in chickpea (7.00 % eggs) followed by soybean (9.50 % eggs) and black gram (12.00 % eggs). In the no choice test also, green gram showed maximum preference for egg laying by *C. maculatus* (87.00 % eggs) followed by cowpea (84.70 % eggs) and pea (83.25 % eggs) which shows similar trend as obtained in choice test. However black gram, soybean and chickpea also showed significantly high egg laying of 81.00 %, 77.00 % and 74.00 %, respectively

The present findings show that the green gram was the most preferred pulse for oviposition by *C. maculatus* while the chickpea was the least preferred pulse. Yunus *et al*. (2015) reported the maximum ovipositional preference of *C. chinensis* in green gram (132.00 eggs) followed by cowpea (78.00 eggs) while the minimum ovipositional preference was recorded on black gram (63.00 eggs) followed by gram (66.67 eggs) which supports our findings. The findings of the present investigation are in partial conformity with the findings of Gurjar (2000) who observed the maximum ovipositional preference in cowpea (71.6 eggs) and the minimum in black gram (50.8 eggs). Similarly, Badoor *et al.* (2009) also reported the ovipositional preference in cowpea which is also in partial conformity with our findings. Radha and Susheela (2014) also reported the maximum ovipositional preference on cowpea (70.14 eggs) followed by black gram (67.14 eggs), green gram (63.38 eggs) and pea (49. 02 eggs). The maximum ovipositional preference by *C. maculatus* was found on pea followed by black gram, chickpea and lentil (Islam *et al.*, 2007) which is slightly similar to the results obtained in the present investigation. Maximum ovipositional preference in cowpea was previously documented (Dwivedi *et al*., 2020; Ahmad *et al.* (2018) also found the maximum number of eggs in cowpea (73.1 eggs) while the minimum number of eggs in black gram (19.5 eggs), also supports our findings. The physical appearance and seed coat affects the oviposition of pulse beetle (de Sa *et al.*, 2014), presumably this might be the reason for more ovipositional preference towards green gram.

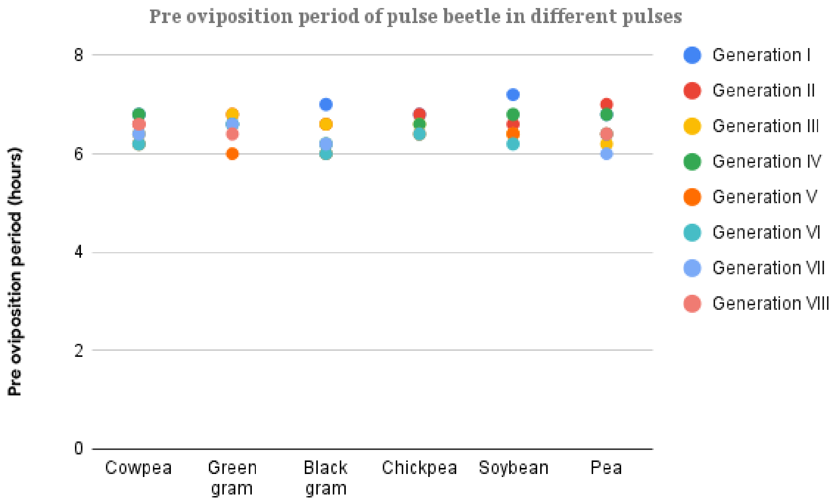


**Fig. 1.** Pre oviposition period of *C. maculatus* in different pulses.

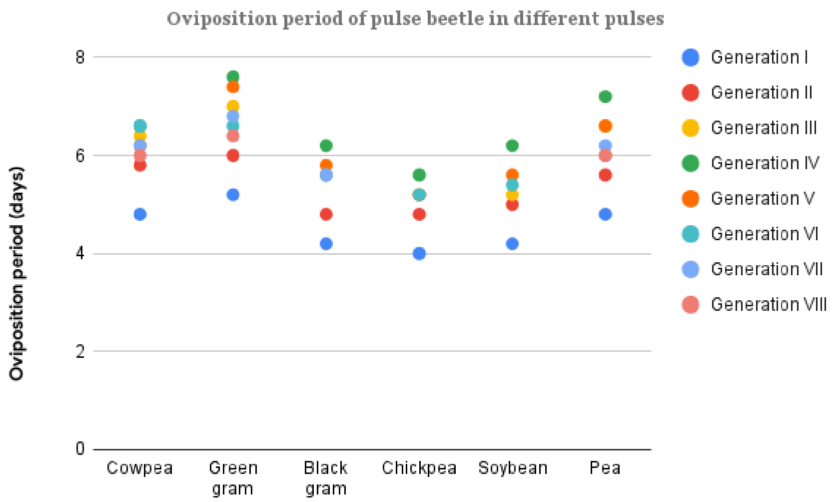


**Fig. 2.** Oviposition period of *C. maculatus* in different pulses.

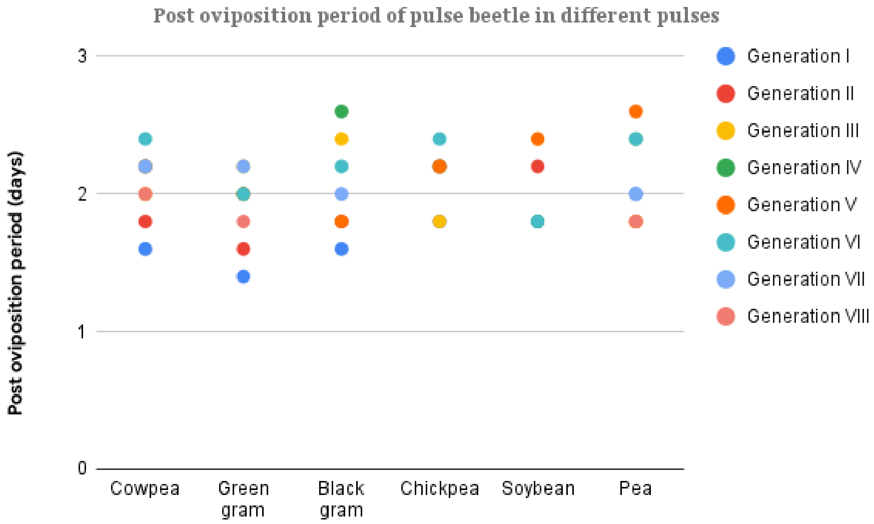


**Fig. 3.** Post oviposition period of *C. maculatus* in different pulses.

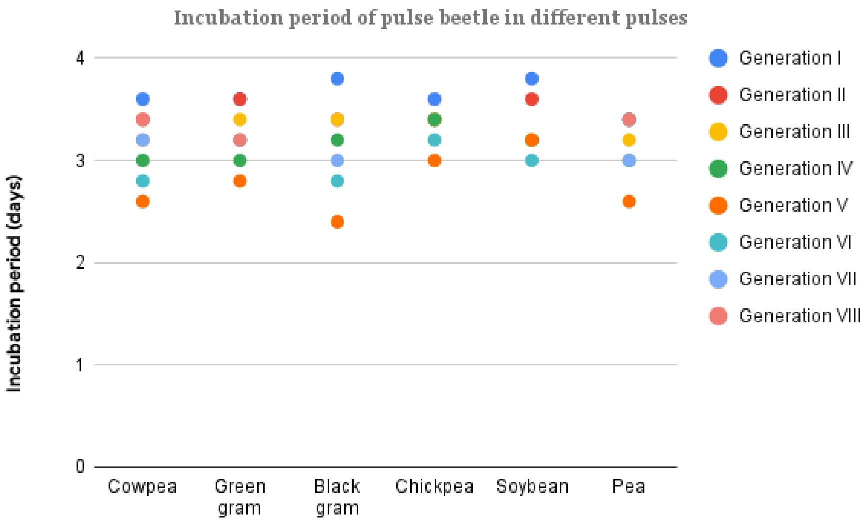


**Fig. 4.** Incubation period of *C. maculatus* in different pulses.

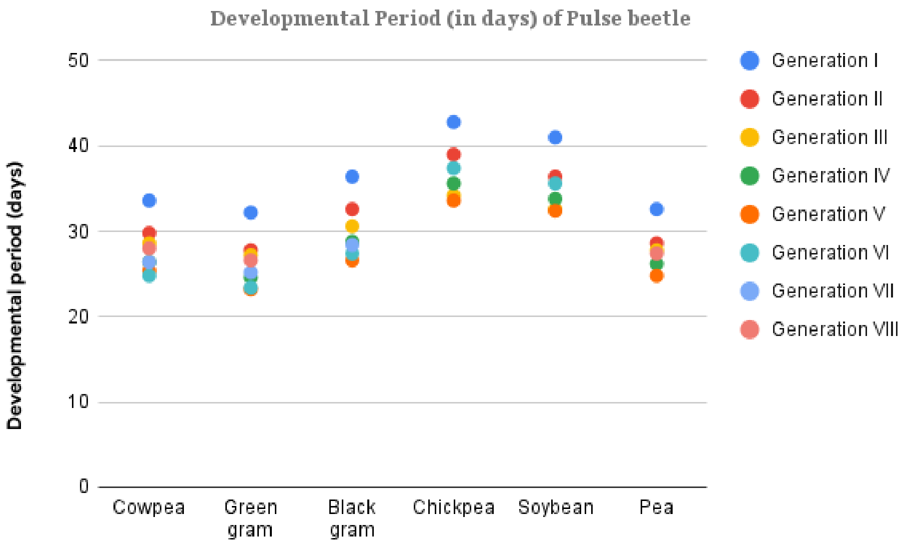


**Fig. 5.** Developmental period of *C. maculatus* in different pulses.

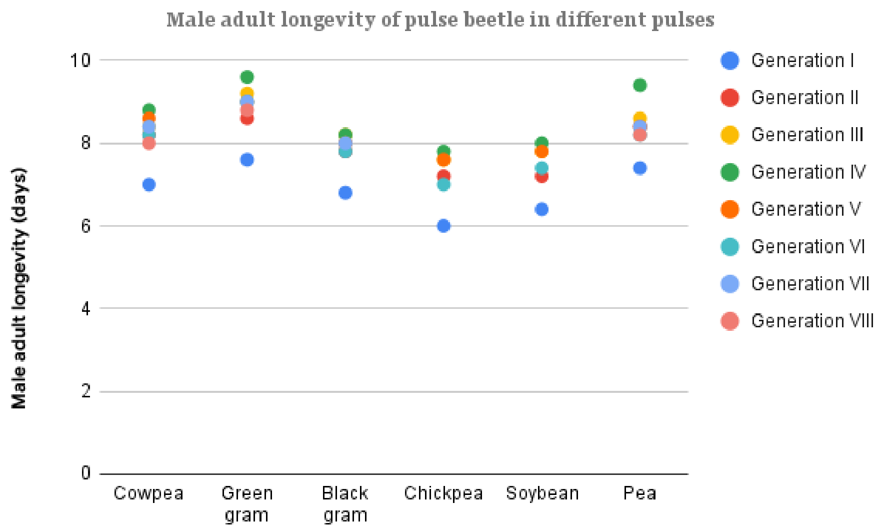


**Fig. 6.** Adult longevity (male) of *C. maculatus* in different pulses.

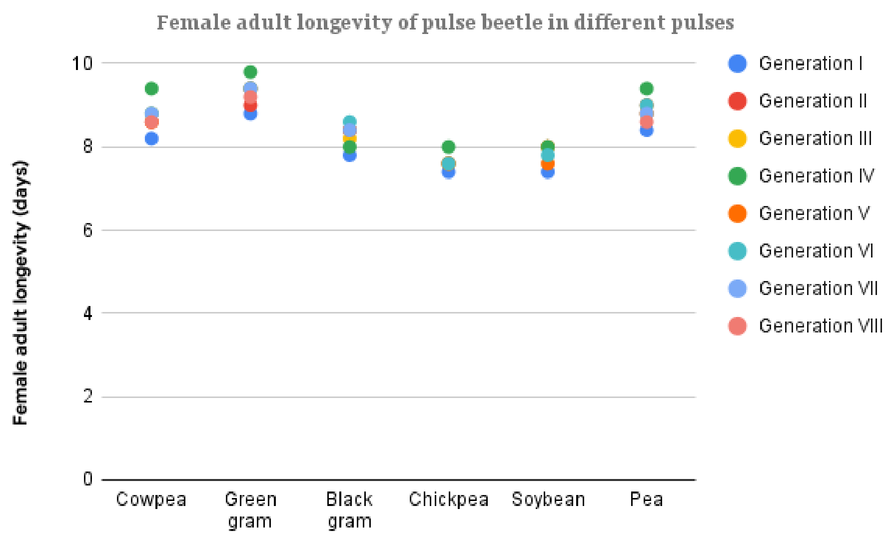


**Fig. 7.** Adult longevity (female) of *C. maculatus* in different pulses.

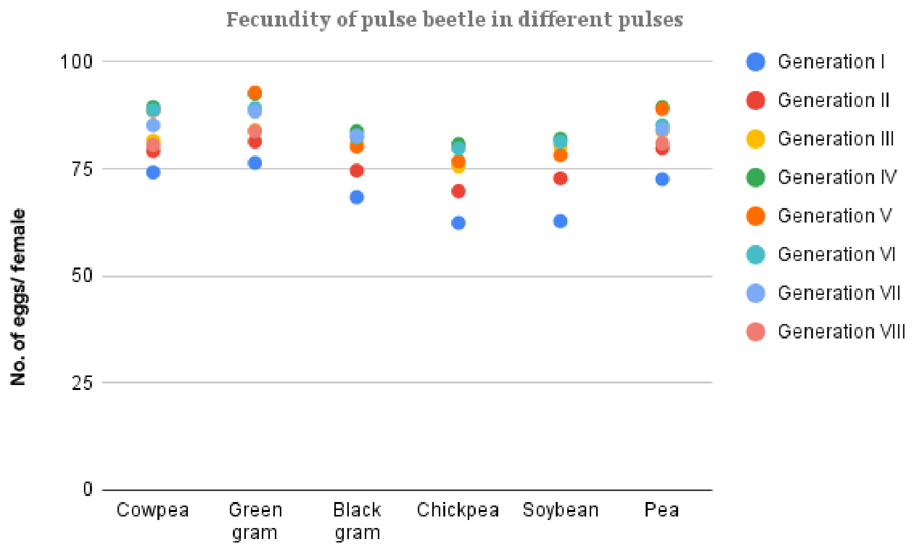


**Fig. 8.** Fecundity of *C. maculatus* in different pulses.

**Table 1: Ovipositional preference of *C. maculatus* in different pulses.**

| Pulses | Free Choice Test | No Choice Test |
|---|---|---|
| **Cowpea** | 16.50[ab] (4.05) | 84.70[ab] (9.21) |
| **Green gram** | 19.25[a] (4.38) | 87.00[a] (9.33) |
| **Black gram** | 12.00[c] (3.46) | 81.00[b] (9.00) |
| **Chickpea** | 7.00[d] (2.62) | 74.00[c] (8.60) |
| **Soybean** | 9.50[cd] (3.07) | 77.00[c] (8.77) |
| **Pea** | 15.75[b] (3.96) | 83.25[ab] (9.12) |
| **C.D** | 0.44 | 0.26 |

Values in parenthesis are square root transformed

## CONCLUSIONS

Our study reveals the generation wise biology of *C. maculatus* in different pulses which is an incipient to better understand the biology of pulse beetle. The life cycle of pulse beetle was quicker in green gram when compared to other pulses. Similarly, the green gram was also found to be the most preferred pulses by *C. maculatus.* As chickpea was found to be the least preferred pulses, it can be concluded that chickpea is resistant to oviposition by *C. maculatus* while green gram is more susceptible to its oviposition.

## FUTURE SCOPE

The molecular level studies should be conducted in future to understand the interaction between the seed coat and biochemical contents of pulses with the biology and oviposition of pulse beetle, which will provide the ways to manage pulse beetle effectively.

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