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Studies on the biology of *Helicoverpa armigera* on different semi-synthetic diet

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Abstract

A study was conducted to determine the effect of artificial diets on some of the important biological parameter of *Helicoverpa armigera* under laboratory condition at 25±1°C, 75±5% R.H. and 14 hr photoperiod. Four locally available pulses viz., chickpea, green gram, pea and black gram were used as treatments and compared with that of the natural diet as control i.e. chickpea leaves. Sixty larvae in each treatment were fed and studied. Results revealed that larval duration was found to be the minimum in larvae reared on chickpea based diet and longer on pea, green gram and black gram respectively and maximum on chickpea leaves. Overall, the fitness index was found highest on chickpea followed by pea, green gram, black gram, black gram and chickpea leaves. The study proved that chickpea based diet was the best for mass rearing of *Helicoverpa armigera* while pea based diet could also be used as substitute.

Keywords: Artificial diet, *Helicoverpa armigera*, laboratory condition, chickpea

Introduction

H. armigera is a major economic pest of many agricultural and horticultural crops (Torres-Villa *et al.*, 1996) [1]. It feeds on over 300 species belonging to 68 plant families around the world, including major crops such as cotton, soybean, maize and a wide range of horticultural crops (Pearce *et al.*, 2017) [2]. Its distribution is expanding and includes at least 145 countries and territories (51 in Africa, 42 in Asia, 29 in Europe, 20 in Oceania, and 3 in South America) (Sullivan and Molet, 2014) [3]. A single larva of *Helicoverpa* has been reported to damage 25-30 pods in its life time (Singh and Ali, 2005) [4]. The worldwide annual costs for controlling this pest along with yield losses reach an estimate of US\$ 5 billion (Lammers and MacLeod, 2007) [5]. According to Hayden and Brambila (2015) [6], the global losses from this pest can be in excess of \$2 billion annually. Considerable damage by *H. armigera* has been reported from almost all major states of India like Andhra Pradesh, Madhya Pradesh, Karnataka, Assam, Punjab, Bihar, West Bengal, Haryana and Gujarat wherein the loss estimates vary from year to year and from crop to crop which is mostly dependent on the pest population density; cotton being highly affected with 80 per cent loss (Fakrudin *et al.*, 2004) [7] followed by 72 per cent in chickpea and in tomato the loss is up to 40 per cent (Setiawati *et al.*, 2000) [8].

The average crop losses in India due to this polyphagous pest are estimated to be US \$350 million annually (Lammers and MacLeod, 2007) [5]. *H. armigera* is a charismatic insect pest in agriculture accounting for the consumption of over 55 per cent of total insecticides used in India (Puri, 1995) [9]. The problem of this pest is magnified due to its direct attack on fruiting structures, voracious feeding habits, high mobility, fecundity and multivoltine overlapping generations (Sarode, 1999) [10].

Moreover, laboratory rearing using synthetic diets is a better option for knowing its biology under controlled conditions. Successful rearing by using synthetic diet becomes a priority to study its life history and various nutritional requirements. Many researchers attempted to rear *H. armigera* under laboratory conditions using synthetic diets (Castane and Zapata, 2005) [11]. The rearing of phytophagous insects on artificial media, rather than on their host plants, is advantageous in a variety of investigations. Laboratory-reared larvae can be used for the study of insect pathogens, plant resistance factors, and effects of insecticides and radiation on fecundity and growth, as well as for the study of insect life cycles (Ahmed *et al.*, 1998) [12]. So, a study was done in College of Post Graduate Studies, (CAU), Umiam, Meghalaya, to study

some of the biological parameters of this insect pest by using some locally available pulses as the main ingredients of the semi-synthetic diet.

Materials and Methods

Laboratory rearing of *H. armigera*

The culture of *H. armigera* was initiated in the laboratory by using pupae procured from the NBAIR, Bengaluru and subsequent generations were established for further study in the research during the year 2017-18. The culture was maintained at $25\pm 1^\circ\text{C}$, $70\pm 5\%$ R.H. and 14 hr light: 10 hr dark (LD 14:10) photoperiod. Larvae obtained from the subsequent generation were used for the study of biology.

The pupae were surface sterilised by 10 per cent sodium hypochlorite solution, sexed and kept in the translucent, cylindrical plastic jars of diameter 12 cm and height 22 cm at the ratio of 5:5 male and female. The perforated holes on the bottom of the plastic jars were covered by blotting paper. It was put into another plastic jar of same size containing some water to maintain the humidity. Adults emerged were fed with 10 per cent honey solution and muslin clothes were covered on the mouth of the jars tighten with rubber bands which served as oviposition sites.

When the eggs hatched out to larvae, they were fed on chickpea based semi-synthetic diet and multiplied. After 2nd instar they were fed individually in the petri dishes of diameter 6 cm to avoid cannibalism.

Description of the diet

The basic composition of the diet was as prescribed by Nagarkatti and Sathyaprakash (1974) [13] modified by Gujar *et al.* (2004) [14]. It consisted of part A and part B. For small scale preparation, part A was made by mixing 84 g of major component (different pulses) with 11 g yeast, 5 g casein, 3 g ascorbic acid, 2 g methyl-p-hydroxybenzoate, 1 g sorbic acid, 0.2 g streptomycin sulphate, 0.2 g cholesterol, 200 µl multivitamin, vitamin E 1 g and 1 ml formaldehyde (10%) in 400 ml of distilled water. Pulses used as major components were chickpea, green gram, black gram and pea. Ingredients in Part A was transferred to a blender and mixed thoroughly for two minutes with 400 ml warm distilled water to get a homogenous mixture. In part B portion, agar was made into 225 ml solution, cooled down to 60°C , and added to Part A. The entire diet was again mixed thoroughly. The homogenous diet was distributed to glass petri dishes (15 cm diameter and 2 cm) and allowed to cool down at room temperature and later stored at 4°C .

Until the second instar, the larvae were fed together on the diet on the petri dishes (6 cm diameter). After that the larvae were reared individually and the diet was renewed every 1-2

days. For comparison, the insect was also fed on the chickpea leaves in similar condition.

Observation recorded

Following observations were recorded:

Larval observations

Larval duration: It was taken from the hatching of eggs till pupation.

Larval weight: It was observed on the fifth instar.

Full size: It was observed on the fifth instar.

Pupal observations

Pupal duration: It was taken from the starting of pupation till adult emergence.

Pupal weight: It was taken few days after pupation.

Female emergence per cent = (No. of female emerged/Total no. of adult emerged) $\times 100$

Male emergence per cent = (No. of male emerged/Total no. of adult emerged) $\times 100$

Growth and fitness index

The larval and pupal growth index and the fitness index were calculated using the following equations (Itoyama *et al.*, 1999) [15]:

Larval growth index = Pupation (%) / Larval period (days)

Pupal growth index = Emergence (%) / Pupal period (days)

Fitness index = [Pupation (%) \times pupal weight] / [Larval period + pupal period]

Statistical analysis

Larval and pupal parameters (duration, weight, size) on different diets were statistically analysed by using one-way analysis of variance (one-way ANOVA) on MS Excel 2016. Fisher's least significance difference (Fisher's LSD) was used to compare pairwise differences between the larvae.

Results and Discussion

Mean values on larval, pupal and adult parameters were observed. (Table 1)

Table 1: Growth and fitness indices

Sl. No.	Treatment	Larval growth index	Pupal growth index	Fitness index
1	Chickpea	7.49	5.47	1.48
2	Green gram	5.71	4.69	1.06
3	Pea	6.31	4.95	1.23
4	Black gram	5.72	4.28	0.93
5	Chickpea leaves (Control)	5.07	3.99	0.80

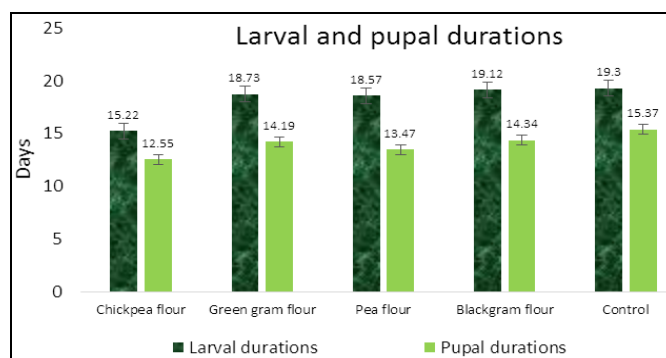


Fig 1: Effects of different semi-synthetic diets on larval and pupal duration of *H. armigera*

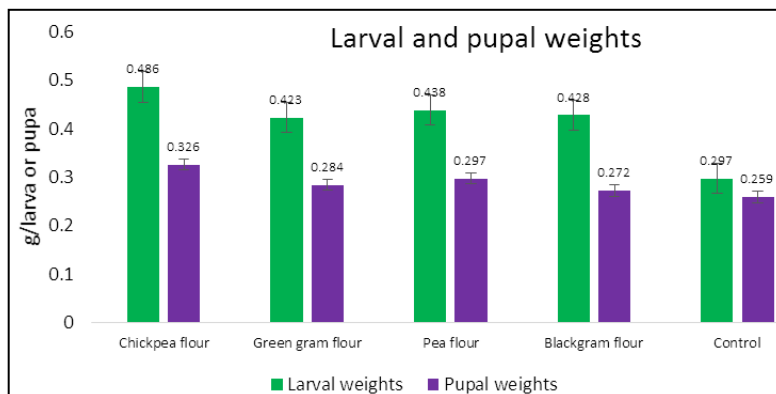


Fig 2: Effects of different semi-synthetic diets on larval and pupal weights of *H. armigera*

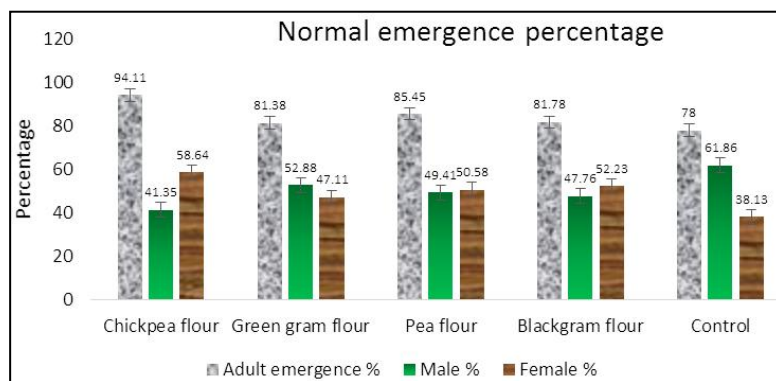


Fig 3: Effects of different semi-synthetic diets on some of the biological parameter of *H. armigera*

Table 2: Mean values (\pm SE) of different biological parameters of *H. armigera* (Hub.) fed on various artificial diets

Sl. No.	Major diet ingredients	Larval			Pupal			Adult		
		Duration (days)	Weight (g/larva)	Full size (cm)	Duration	Weight (g/pupa)	Pupation (%)	Normal emergence (%)	Male (%)	Female (%)
1	Chickpea flour	15.22a (\pm 0.28)	0.486a (\pm 0.007)	3.42a (\pm 0.11)	12.55a (\pm 0.92)	0.326a ^{#*} (\pm 0.012)	95.58a (\pm 0.40)	94.11a (\pm 0.43)	41.35a (\pm 0.75)	58.64a (\pm 0.75)
2	Green gram flour	18.73b (\pm 0.19)	0.423b (\pm 0.006)	3.27a (\pm 0.09)	14.19a (\pm 1.12)	0.284b (\pm 0.015)	88.33b (\pm 0.51)	81.38b (\pm 0.81)	52.88b (\pm 0.83)	47.11b (\pm 0.83)
3	Pea	18.57b* (\pm 0.20)	0.438b (\pm 0.004)	3.27a (\pm 0.11)	13.47a (\pm 0.93)	0.297b ^{^#} (\pm 0.009)	91.67c (\pm 0.81)	85.45c (\pm 0.82)	49.41c (\pm 0.98)	50.58c (\pm 0.98)
4	Black gram flour	19.12b (\pm 0.16)	0.428b (\pm 0.021)	3.35a (\pm 0.05)	14.34a (\pm 0.86)	0.272b* (\pm 0.005)	81.67d (\pm 1.04)	81.78b (\pm 0.99)	47.76c (\pm 0.94)	52.23c (\pm 0.94)
5	Chickpea leaves (control)	19.30b* (\pm 0.14)	0.297c (\pm 0.005)	2.89b (\pm 0.06)	15.37a (\pm 0.92)	0.259b [^] (\pm 0.006)	77.14e (\pm 0.97)	78.00b (\pm 1.10)	61.86d (\pm 1.11)	38.13d (\pm 1.11)
SEM \pm		0.75	0.031	0.09	0.47	0.01	3.34	2.76	3.37	3.37
CD at 5%		0.62	0.033	0.27	NS	0.03	2.44	2.68	2.90	2.90

Means sharing same letters are statistically non-significant, but those with similar symbols are significant, ($P < 0.05$, LSD).

It was found that the larval duration was the shortest on the chickpea-based diet (15.22 days) which was followed by pea (18.57 days), green gram (18.73 days) and black gram (19.12 days) and the longest on chickpea leaves (control) (19.30 days) (Fig.3). Mean larval weight (Table 2) was found to be the highest on chickpea diet (0.486 g/larva) followed by pea, black gram, green gram and control i.e., 0.438 g, 0.428 g, 0.297 g per larva, respectively. Mean pupal weight (Table 2) was the highest on chickpea diet (0.326 g/pupa) followed by pea, green gram, black gram and control i.e., 0.297, 0.284, 0.272, 0.259 g per larva respectively. Pupation percentage (Table 2) was found to be the highest in chickpea (95.58%), followed by pea (91.67%), green gram (88.33%), black gram (81.67%) and chickpea leaves (77.14%) respectively. Emergence of normal adults (Table 2) was found highest on chickpea-based diet (94.11%) followed by

pea (85.45%).

The finding revealed that more per cent emergence of female was produced in chickpea-based diet (58.64%) which was followed by black gram (52.23%), pea (50.58%), green gram (47.11%) and chickpea leaves (38.13%). Larval growth index (Table 1) was found to be the highest on chickpea (7.49) followed by pea (6.31), black gram (5.72), green gram (5.71) and control (5.07). Similarly, pupal growth index on chickpea (5.45) was the highest followed by pea (4.95), green gram (4.69), black gram (4.28) and control (3.99). The fitness index on chickpea (1.48) showed the highest followed by pea (1.23), green gram (1.06), black gram (0.93) and control (0.80). This indicated that chickpea based diet could also be used as substitute.

According to present findings, chickpea-based diet was noticed to be the most suitable for mass rearing of *H.*

armigera. Our results are in conformity with the findings of Amer and El-Sayed (2014) ^[16] who reported that the developmental periods of larvae of *H. armigera* was the shortest on pea-based artificial diet (17.50 days). Similar results were also observed by Nunes *et al.* (2017) ^[17] who reported that artificial diets were more adequate for *H. armigera* and the chickpea-based diet resulted in a shorter generation time of *H. armigera*. The present results more or less agree with the study made by Pimparkar and Raja (2017) ^[18] who reported that body growth, rate of maturity and number of pupa and adult formed were the highest as well as mortality rate and time taken for growth were the least in larvae fed on artificial diet.

Our findings highlighted that the larval duration, mean larval weight, mean pupal weight, adult emergence and female emergence percentage are 15.22 days, 0.486 g, 0.326 g, 94.11% and 58.64%, respectively. Our findings are in close agreement with Hamed and Nadeem (2008) ^[19] who reported the above values as 14 days, 0.45 g, 0.38 g, 91.6% and 77%, respectively. But Nunes *et al.* (2017) ^[17] reported the larval duration on chickpea-based diet (15.22 days) was 11 days which is shorter from our present findings. The probable reason may be the difference in the temperature as they reared the culture at higher temperature i.e., at 28±2 °C.

Conclusion

The shortest time period was taken by the larvae reared on the chickpea-based diet followed by pea, green gram, black gram and control respectively. Chickpea based diet exhibited better performance on other biological parameters too. Thus, fitness index on chickpea (1.48) showed the highest followed by pea (1.23), green gram (1.06), black gram (0.93) and control (0.80). Pea could also be used as a substitute for chickpea.

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