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EFFECT OF REARING METHODS ON *SPODOPTERA LITURA* (F) UNDER LABORATORY CONDITION

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Effects of natural and artificial diets under two rearing methods (group and individual) on larval and pupal development and survivability of *Spodoptera litura* (Fab.) were studied. The survival rate of larvae and pupae was significantly affected by diets (P = 0.05), which was higher on artificial diet when reared individually. Results of this study documented the higher larval and pupal growth index from artificial diet. The larval growth indices were 5.24 and 1.65 on artificial diet under individual group rearing, respectively, and 2.35 on natural diet under group rearing. The developmental periods of larvae and pupae were not significantly affected by diets, which were shorter on artificial diet reared in group.

Key words: *Spodoptera litura*, artificial diet, rearing methods, adult emergence, reproductive competence, larval growth index, pupal growth index, larval survival, pupal survival

Spodoptera litura (Fab.) is a polyphagous species widely distributed throughout the world due to its migratory behavior (Rani and Mohan, 2000; Narayanan, 2003). Around 112 plant species belonging to 44 families have been reported as its food plants (Xue et al., 2010). The pest is capable of eating leaf, flowers, fruit etc. and 26 to 100% losses have been documented depending upon the crop stage and its infestation level in the field (Daniel and Samiayyan, 2017). Rearing of an insect species under artificially controlled conditions is the pre-requisite for its continuous supply, for conducting various experiments regarding evaluation of toxicity and efficacy of insecticides. Diet and rearing methodology plays a critical role on the growth and development, survival, adult behavior, and reproductive competence of reared insect. Literature revealed that different workers used different rearing regimes for larval rearing of *S. litura* like rearing larvae in group on natural diet or artificial diet and rearing individual larvae in separate vials, but, the studies on comparison of different rearing regimes of *S. litura* are lacking. Therefore, present study has been conducted to compare the effects of rearing methods on growth index and survival and survivability of *S. litura*.

MATERIALS AND METHODS

The present investigation was carried out at ICAR-NRCIPM and Division of Entomology, ICAR- IARI, Pusa Campus, New Delhi. The experiment was conducted in a growth chamber (Accumax India) during 2018-19. Fourth and fifth instar larvae of *S. litura* were collected from the fields of ICAR-IARI, in 2018, and reared on fresh castor leaves. The culture was maintained at 27± 1ºC and 65± 5 % RH with a 14:10 L: D photoperiod. The experiment was conducted with four treatments i.e. artificial diet- individual (ADI) artificial diet in group (ADG) group rearing on natural diet i.e. castor leaf (NDG) (N.D.G) and individual rearing on natural diet (NDI).

Group and separate larval rearing on natural diet was done by providing fresh castor leaves. Neonates (<12 hrs) were transferred to sterilized glass container (22 cm x 14 cm) which had a small cup half-filled with tap water where petioles were submerged to prevent the desiccation of leaf and about 30-40 neonates were released in each of the two leaves maintained per container. Fresh castor leaves were sterilized by 1% sodium hypochlorite solution followed by washing with tap water about 3-4 times before feeding in every alternate day. After 5th day of hatching in group rearing, 10 larvae were transferred to each propylene jar (20 cm x 15.5 cm) whereas for separate rearing the larvae were individually placed in small rectangular vials (5 cm x 2.5 cm x 4.5 cm). Survivability was recorded at every 24 hr until the emergence of healthy adult. Pre-pupae were allowed to pupate inside jar till completion of sclerotization and pupae were transferred to new sterilized petri plates for adult emergence.

Group and individual larval rearing on artificial diet was done by providing chickpea flour based diet, prepared as per the protocol reported by Gupta et al.,

2005[a] with some modifications. Neonates were reared in the same manner as described above on castor leaves. After 5th day of hatching, the survived larvae were individually placed in small rectangular vials for individual rearing and 10 larvae for group rearing were placed in a Petri plate (16 x 3 cm). Larvae were allowed to pupate inside the artificial diet till 48 hr and shifted from diet to vials for adult emergence.

Observations were recorded on some attributes of growth and development of the immature stages of *S. litura*. Since NDI resulted inconsistent feeding by larvae and malformation in pupae, poor quality of malformed pupae, it was excluded from data analysis of growth indices. One day old neonates were used to measure the biological characteristics, selected randomly from the same egg patch, larvae were weighed on 8th, 14th and 17th day of hatching, respectively which was standardized as optimal time to measure the diet accessibility and growth (Sridhar et al., 2013). Only healthy, live pupae were weighed and counted. The larvae with unsuccessful moulting and adult with malformed organs were considered dead. The larval and pupal growth indices were calculated using the following formulae (Gupta et al., 2005[b]): Larval growth index= % pupation/ larval duration (days); Pupal growth index= % of emerged moths/ pupal duration (days); Larval period (days), larval survivability (%), larval weight (mg), pupal period (days), pupal weight (mg), rate of pupation (%) and rate of adult emergence (%) were recorded. Each treatment was replicated three times and ten larvae per replication. The experiment was laid out in completely randomized design with 4 treatments and 3 replications; data analysis was done by using WASP 2.0 package.

RESULTS AND DISCUSSION

Mean percentage of survival of 1st and 2nd instar larvae of *S. litura* were at par in all the treatments (Table 1). Survivability of 3rd instar larvae was significantly higher in ADI (98.05%) followed by ADG and NDG. Similarly survival of 4th, 5th and 6th instar larvae in ADI was significantly higher followed by NDG and ADG. Consequently pre-pupation rate was also observed highest on ADS (93.61%) followed by NDG (51.12%) and lowest on ADG (38.47%). The duration of 1st, 2nd and 4th instar larvae and prepupal stage was statistically at par in all the treatments. Duration of 6th instar was lowest in ADI (1.15 days) followed by ADG and NDG. Total larval period was minimum in ADI (16.68 days) followed by ADG and NDG. The mean larval weight was significantly higher in ADI. Pupation was significantly higher on ADI (89.44%), followed by NDG and ADG. The pupal periods were at par in all treatments. Mean pupal weight was significantly affected by different treatments which was maximum in ADI (323 mg) followed by ADG and NDG. Rate of adult emergence was also highest in ADI (86.8%) followed by NDG and ADG. ADI recorded the highest larval growth index (5.24) and pupal growth index (11.42) followed by NDG and ADG (Table 1).

According to Bernays and Chapman (1994) growth, development, survival and reproduction in phytophagous insects significantly affected by the chemical composition of diet and development of larvae is influenced by the food quality. In this study, we have examined the effect of different diets on larvae development. Although, a long list of different artificial diets have been reported for the culturing and maintenance of the such economically important insect pest, generally for lepidopteran larvae (Cohen, 2001) but there is always a chance of the refinement of existing standard protocols. The lowest mean survival rate of larvae and pupae on natural food indicates that castor leaves failed to supply required nutrients which were essential to change the consumption rate into different metabolic reactions. Chickpea was the main source of carbohydrate (Seth and Sharma, 2002), to avoid the chances of fungal and other secondary infections, microbial inhibitors were mixed during the preparation of diet for example streptomycin, sorbic acid, methyl-p-hydroxybenzoate (methyl paraben). L-ascorbic acid used as indirect source of vitamin-c, which is an essential requirement for normal growth of herbivores. The study showed low survival of *S. litura* larvae under group rearing condition on artificial diet and natural diet in comparison to separate rearing on artificial diet, which may be due to the cannibalism among the larvae because of limited space and competition for food. According to Bloom et al. (2010) insects were very sensitive to any type of stress (ecological and biological) to their surroundings, that can totally change their growth and development. Previous studies reported that non motile (egg and pupae) or less motile stages (younger larvae) are more prone to cannibalism by older larvae and more on natural host when compare to artificial diet in mass rearing. Ryuda et al. (2008), favored to rear larvae separately on artificial diet to avoid cannibalism in tobacco cutworm. Separate or singly rearing on artificial diet in vials, is a good option to avoid cannibalism in laboratory rearing. Janakiraman and Gupta (2002) reported 86.2% pupation, 325.9mg

Table 1. Survival, growth and development of *S. litura* on artificial diet and castor leaf

Treatment	Different stages of *S. litura*					
	3rd Instar	4th Instar	5th Instar	6th Instar	Pre- pupa	Pupa
ADI	98.05 (85.14)[a]	97.23 (83.10)[a]	96.39 (81.06)[a]	94.16 (77.67)[a]	93.61 (77.01)[a]	89.44 (72.28)[a]
ADG	86.69 (66.43)[b]	73.2 (59.79)[c]	58.05 (49.79)[c]	48.89 (44.33)[c]	38.47 (38.17)[c]	29.16 (32.50)[c]
NDG	85.55 (68.22)[b]	78.33 (62.72)[b]	73.33 (59.38)[b]	60.03 (50.56)[b]	51.12 (45.63)[b]	37.5 (37.62)[b]
S.Em	0.087	0.076	0.08	0.105	0.055	0.1
CD (p=0.05)	7.89	7.94	5.91	5.05	5.44	5
Treatment	3rd Instar	4th Instar	5th Instar	6th Instar	Pre- pupa	Pupa
ADI	3.54 (1.91)[a]	4.075 (2.019)[a]	2.37 (1.82)[a]	1.15 (1.59)[a]	1.02 (1.24)[a]	7.6 (2.67)[a]
ADG	3.13 (1.8)[a]	4.04 (2.061)[a]	2.32 (1.69)[a]	1.38 (1.67)[a]	1.44 (1.37)[a]	7.3 (2.87)[a]
NDG	2.78 (0.707)[b]	4.42 (1.761)[b]	2.2 (1.52)[a]	2.47 (0.707)[b]	1.47 (1.17)[a]	8.28 (3.05)[b]
S.Em	0.080	0.059	0.084	0.066	0.048	0.103
CD (p=0.05)	0.11	0.167	NS	0.11	NS	0.197
CV	3.26	3.42	5.41	11.93	10.56	3.94

Growth and development (Mean± S.Em)

Parameter	T1	T2	T3
Larval weight (mg) (8-day-old)	50.06± 1.23	49.58± 1.85	48.17± 2.6
Larval weight (mg) (14-day-old)	428.04± 34.07	312.64± 27.74	122.18± 10.01
Larval weight (mg) (17-day-old)	742.15± 63.45	608.76± 4.70	279.44± 61.25
Larval period (days)	17.06± 0.49	17.88± 0.55	15.9± 0.35
Pupal period (days)	7.6± 0.15	7.3± 0.19	8.28± 0.18
Pupal weight (mg)	323± 19.77	249± 11.29	190.52± 26.92
% of Emerged Adults	86.8± 0.57	28.01± 4.34	34.13± 3.66
Larval growth Index	5.24	1.65	2.35
Pupal growth Index	11.42	3.83	4.12

ADI artificial diet; ADG: artificial diet group; NDG: group rearing natural host; *Figure in parenthesis are arc sign transformed values; values superscript with same letter are statistically *at par*. Artificial diet-individual; ADG: artificial diet group; NDG: group rearing on natural host; Figures in parenthesis are square root transformed values and values superscript with same letter are statistically *at par*.

pupal weight, 92.3% adult emergence and 83.8% survival rate, for the same insect when reared on artificial diet. Narvekar et al. (2018) reported 7.33 days pupal period on castor, however, it was found to be longer (8.28 days) on castor in our study. Our results on artificial diet in separate rearing indicating highest larval and pupal growth index which seem to be co-incide with the results by Divakara and Manjulakumari, (2015). It can be concluded that the group rearing of *S. litura* whether on artificial or natural host (castor leaf) is not feasible in the laboratory conditions. Individual rearing on artificial diet is a good alternative to group rearing on natural/ artificial diet.

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AUTHOR CONTRIBUTION STATEMENT

All authors equally contributed.

CONFLICT OF INTEREST

No conflict of interest.

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