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Rearing of *Helicoverpa armigera* (Hub.) on Artificial Diets in Laboratory

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Abstract.- Seven artificial diets were prepared by substituting basic ingredients as flour of chickpea, mungbean, soybean, wheat, maize, cotton seed and water chestnut and tested for rearing *Helicoverpa armigera* in laboratory and compared with natural food comprising of chickpea leaves and pods. Chickpea flour mediated diet produced healthy larvae and pupae that gained the maximum weight measuring 0.4500 and 0.3805 g and completed development within the minimum duration of 14.5 and 11 days, respectively. Flour of water chestnut reduced their weight to 0.2742 and 0.1775 g with the increased developmental duration of 42.8 and 14.8 days, respectively, as compared to flour of cotton seed, maize and wheat. Likewise, the larval length ranged from 3.7 to 3.3 cm in all treatments. Mortality of larvae was maximum on flour of cotton seed followed by flours of water chestnut, soybean and wheat. However, mortality of *H. armigera* was minimum and non significant ranging from 1.1 to 1.3% in flour of chickpea, mungbean and natural chickpea leaves and pods. Pupal recovery ranged from 60.3 to 80.1%, with maximum on chickpea flour, and minimum on water chestnut diet. Percent male and female emergence was changed accordingly in all treatments. Apparently normal adult emergence of 91.6% was achieved on chickpea flour, 85.5% on mungbean, 83.1 % on chickpea leaves and pods and 82.5% on soybean. It was concluded that chickpea flour mediated diet along with other essential ingredients are very conducive to get good quality culture of *H. armigera*. Mungbean and soybean were the other main ingredients of diets that can be used for successful culture rearing.

Key words: Artificial diets**,** *Helicoverpa armigera*, insect rearing.

INTRODUCTION

Helicoverpa armigera (Hub.) is a major pest of cotton, chickpea, tomato, okra and some other crops and vegetables in Pakistan (Ahmad *et al*., 1989). It has been recorded to damage 60 cultivated plant species and at least 67 other plant species in 39 families across the world (Reed and Powar, 1982). To control its wide existing population, many methods are being applied that include insecticides, biological control, pheromones, host plant resistance, mechanical and genetically modified crops. The above methods of control can be successful after careful studies of different biological parameters of this injurious pest in the laboratory. Bioassay is an important technique that is being in practice in laboratories for pesticide and *B.t.* toxin evaluations against this pest. For bioassay, the year around availability of this insect pest is imperative. Laboratory reared larvae can also be used for the study of insect pathogens, plant resistance factors, effects of insecticides and radiations on fecundity and growth and its own biology. To meet the demand of laboratory reared insects low cost artificial diets are required that could improve their development. The essential ingredients for insect development of artificial diets includes proteins, carbohydrates, fats, cholesterol, glucose, vitamins, minerals and preservatives etc. Singh and Rembold (1988) reported that rate of survival and development period of *H. armigera* larvae were different depending upon diet ingredients *i.e*., chickpea, soybean or maize. Environmental conditions, nutritive and feeding preference may also affect the intake of food by larvae. Successful rearing of *Heliothis zea* on artificial diet has been reported based on soybean flour/wheat germ diet by studying different developmental parameters of insects growth stages (Burton and Perkins, 1972; Vanderzant *et al*., 1962). Ahmad *et al*. (1998) have observed rearing of *H. armigera* on modified artificial diet based on bean powder (*Vigna unguiculata*) and concluded that

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pupal recovery percentage ranged from 71.2 to 83.7% and adult emergence varied from 59.6 to 78.4%. Abbasi *et al*. (2007) has successfully reared *H. armigera* on tapioca based artificial diet and got successful results on developmental parameters of insect during larval stage, larval and pupal developmental period, percent pupation, pupal weight and emergence rate of male and female. Singh (1999) compared feeding of artificial diet prepared with maize, soybean and chickpea and reported that food consumption and growth of *H. armigera* larvae were minimal on maize. Nutritive value of soybean diet was higher but consumption rate of larvae were more on chickpea diet as compared to others.

The present study was undertaken to evaluate the effect of commonly available, easily grinded and low cost flour of chickpea, mungbean, soybean, wheat, maize, cotton seed and water chestnut along with other essential diet ingredients on the development parameters of *H. armigera* larvae under laboratory conditions.

MATERIALS AND METHODS

Present experiment was conducted in beneficial insect rearing laboratories of Plant Protection Division at Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan. Laboratory temperature was maintained at 25±2°C, 75±5% RH and 14-10(L:D) h photoperiod. Fresh larvae of *Helicoverpa armigera* were collected from chickpea crop for their culture maintenance through many generations on natural chickpea leaves and pods. Emerged adults were paired to get eggs and neonate larvae. Seven artificial diets were prepared with the change of basic ingredients including flour of chickpea (*Cicer arietinum* L), mungbean (*Vigna radiata*), soybean (*Glycine max*), wheat (*Triticum aestivum* L), maize (*Zea mays*), cotton seed (*Gossypium hirsutum*) and water chestnut (*Trapa bispinosa*) each measuring 600 g and mixed in a common mixture containing the following three parts:

Part A includes yeast powder (60 g), sucrose (60 g), formaldehyde 10% (15 ml), choline chloride 20% (30 ml), distilled water (1200 ml). Part B includes ascorbic acid (12 g), methyl 4 hydroxy benzoate (7.5 g), sorbic acid (4.5 g), streptomycine sulphate (0.1 g), cholesterol (0.6g), wheat germ oil (0.6 ml) and vitamin mixture (0.6 g). Part C includes agar (45 g) and distilled water (1000 ml). The vitamin mixture in part B contains nicotine acitamide (9.30 g), riboflavin (4.64 g), pyridoxine hydrochloride (2.32 g), biotin (0.18 g), vitamin B_{12} (0.01 g), folic acid (4.64 g) and thiamine hydrochloride (2.32 g).

Preparation of diet

Part C was boiled in distilled water and a homogenous mixture was prepared. Then mixed the ingredients of part A and part B separately and blended in a grinder. Ingredients of part A were poured in to the ingredients of part B and blended and mixed in the ingredients of part C and then kept at room temperature for some time and stored at low temperature (8°C) in refrigerator.

First instar larvae were taken from the stock culture and released in all treatments including control and seven artificial prepared diets, each with six replicate and each replicate was maintained ten larvae. In control larvae were fed on natural chickpea leaves and pods. In each replicate of treatment fifty larvae were released. Daily data on parameters regarding larval duration, weight, length and percent mortality, pupal duration and weight, percent pupal recovery, male and female percentage and percent emergence were recorded. Data was statistically analyzed following Steel and Torrie (1980) and significance was tested by using DMRT.

RESULTS AND DISCUSSION

Results in Table I showed that all insect life variables differed significantly among the treatments. Larval duration of *H. armigera* was observed minimum (14.5 days) in chickpea flour diet that was prolonged to the maximum (42.8 days) in water chestnut flour diet. In other diets larval duration were observed as 15.3, 15.5, 15.6, 16.0, 16.5 and 21.3 days on mungbean, soybean, chickpea leaves and pods (control), wheat, maize and cotton seed respectively. Significantly maximum weight per larva (0.4500 g) was recorded on chickpea diet and minimum weight (0.2742 g) was observed on water chestnut diet. Other statistically comparable

treatments for maximum larval weight was on soybean (0.4438 g), mungbean (0.4276 g) and

Table I.- Mean values (± SE) of different biological parameters of *H. armigera* (Hub.) fed on various artificial diets.

Main diet ingredients	Larvae				Pupae			Adults		
	Duration (days)	Weight (g/larva)	Full size (cm)	Mortality (%)	Duration (days)	Weight (g/pupa)	Pupal recovery (%)	Males (%)	Females (%)	Normal emergence (%)
Chickpea leaves & pods (control)	15.6 ± 0.49a	0.2767 ± 0.00a	2.9 ± 0.06c	1.3 ± 0.49a	14.6 ± 0.42b	0.2599 ± 0.00cd	80.1 ± 1.06c	81.9 ± 1.06a	18.0 ± 1.06f	83.1± 0.6bc
Chickpea flour	14.5 ± 0.09a	0.4500 ± 0.00a	3.3 ± 0.04a	1.1 ± 0.40a	11.0 ± 0.57a	0.3805 ± 0.00a	90.6 ± 0.69a	22.3 ± 0.37f	77.6 ± 0.37a	91.6 ± 0.66a
Mungbean flour	15.3 ± 0.49a	0.4276 ± 0.00ab	3.3 ± 0.04ab	1.3 ± 0.49a	13.0 ± 0.22b	0.3576 ± 0.00ab	85.3 ± 0.50b	34.7± 0.26e	65.2± 0.23b	85.5± 0.84b
Soybean flour	15.5 ± 0.56a	0.4438 ± 0.02a	3.2 ± 0.06b	15.3 ± 0.42b	13.5 ± 0.34b	0.3376 ± 0.00b	76.1 ± 0.44d	34.3 ± 0.39e	65.6± 0.40b	82.1± 0.70c
Wheat flour	16.0 ± 0.36a	0.3926 ± 0.01b	3.1 ± 0.02b	15.3 ± 0.42b	14.5 ± 0.43b	0.2970 ± 0.01c	75.0 ± 0.44d	62.9 ± 0.90c	37.0 ± 0.90d	81.8± 0.79c
Maize flour	16.5 ± 0.22a	0.4032± 0.00b	3.2 ± 0.06ab	18.3 ± 0.66c	14.0 ± 0.51b	0.2688 ± 0.01cd	74.6 ± 0.40d	61.8 ± 0.52c	38.1± 0.52d	70.6± 0.49d
Cotton seed flour	21.3 ± 0.49b	0.2907 ± 0.00c	2.7 ± 0.04b	21.0± 0.51d	14.8 ± 0.40b	0.2568 ± 0.00cd	65.0 ± 0.33e	76.8 ± 0.35b	23.1± 0.35e	61.1± 0.87e
Water chestnut flour	42.8± 1.28c	0.2742 ± 0.00c	2.8 ± 0.06cd	15.8 ± 0.42e	14.8 ± 0.43b	0.1775 ± 0.00e	60.3 ± 0.30f	55.4± 0.90d	44.5 ± 0.53c	55.3± 1.59f

Means sharing same letters are statistically non-significant ($P < 0.05$).

chickpea leaves and pods (0.2767 g) followed by maize (0.4032 g) and wheat flour (0.3926 g). Weight of larva on cotton seed flour (0.2907g) was very close to water chestnut. Length of full grown larva (5^{th} instar) was 3.3 cm on chickpea flour diet followed by 3.3, 3.2, 3.2, 3.1 in mungbean, soybean, maize and wheat flour, respectively. Significantly minimum larval length (2.7 cm) was recorded on cotton seed flour followed by water chestnut (2.8 cm). The minimum mortality was observed on chickpea flour (1.1%) while maximum mortality was on cotton seed flour (21.0%). Mortality percentage on control and mungbean was close to chickpea flour, whereas that of soybean was equal to wheat flour diet, maize flour and water chestnut followed cotton seed. Pupal duration was shorter on chickpea flour (11.0 days) compared with other diets. While pupal period was prolonged to 14.8 days in both cotton seed and water chestnut. However all treatments except chickpea flour behaved non significantly in pupal duration. Pupal weight was 0.3805 g on chickpea flour diet that was highest among all diets. The minimum weight per pupa (0.1775 g) was recorded on water chestnut diet. Weight of pupa in all other diets ranged from 0.2568 to 0.3576 g. Maximum pupal recovery (90.6%) was observed in chickpea diet whereas, minimum (60.3%) on water chestnut. Artificial diets yielded more females than natural diets. In chickpea mediated diet, significantly higher female emergence (77.6%) was observed as compared to male (22.3%) followed by mungbean (65.2% females, 34.7% males) and soybean mediated diet (65.5% females, 34.3% males). The minimum female emergence (18.0%) was recorded in chickpea leaves and pods (control). There was a higher percent normal emergence of adults in chickpea natural (91.6%) and artificial diets (83.1%). Whereas, all other diets showed intermediate effect on moth emergence. The water chestnut diet decreased male adult emergence to 55.3%. The results presented here agree partially with those of previous studies due to variations in the test insect strain and basic diet ingredients used . Brewer and King (1979) have succeeded in rearing *Heliothis* larvae on artificial diet based on soybean flour and wheat germ diet after studying different developmental parameters of the insect growth stages. Ahmad *et al*. (1998) tested the rearing of *H. armigera* on a modified artificial diets and recorded pupal recovery ranging from 71.2 to 83.7% and adult emergence from 59.6 to 78.4%. This finding

was in agreement to our results on adult emergence that ranged from 60.3 to 90.6%. Singh (1999) conducted studies on feeding of artificial diet with main ingredients as maize, soybean and chickpea along with other essential ingredients and found minimal food consumption and growth of *H. armigera* larvae on maize. Nutritive value of soybean diet was high but consumption rate of larvae was more on chickpea diet as compared to others. These results agree with our findings that chickpea diet proved to be a good artificial diet in terms of consumption and development of larvae. Our findings on normal adult emergence are in the line of previous work who got normal adult emergence after feeding larvae on artificial diet (Burton, 1970). So it was concluded from present findings that chickpea flour mediated diet along with other ingredients to be proved more effective for rearing *H. armigera* in the laboratory.

REFERENCES

ABBASI, B.H., KHALIQUE, K., KHALIQUE, F., AYUB, N., LIU, H.J., KAZMI, S.A.R. AND NAUMAN, M., 2007. Rearing the cotton bollworm, *Helicoverpa armigera* on a tapioca based artificial diet. *J. Insect Sci.*, **7**: 35.

AHMAD, K., KHALIQUE, F. AND MALIK, B.A., 1989. Studies on *Helicoverpa armigera* management in Pakistan. *Proc. 1st Int. Conf. Econ. Ent*., **2**: 153-161.

AHMAD, K., KHALIQUE, F. AND MALIK B.A., 1998. Modified artificial diet for mass rearing of chickpea pod borer, *Helicoverpa armigera* (Hub.). *Pakistan J. biol. Sci*., **1**: 183-187.

BREWER, F.D. AND KING, E.G., 1979. Consumption and utilization of soyflour-wheat germ diets by *Heliothis* spp. *Ann. ent. Soc. Am*. **72**: 415-417.

BURTON, R.L., 1970. A low cost artificial diet for the corn earworm and fall armyworm. *J. econ. Ent.*, **63**: 1967-1970.

BURTON, R.L, AND PERKINS, W.D., 1972. WSB a new laboratory diet for corn earworm and the fall armyworm. *J. econ. Ent.*, **65**: 385-386.

REED, W. AND POWAR, C.S., 1982. *Heliothis*: a global problem. In: *Proceeding of the International Workshop on* Heliothis *Management*, 15-20 November ICRISAT, India.

SINGH, A.K., 1999. Growth and induction in food consumption of *Helicoverpa armigera* Hbn. (Lep., Noctuidae) larvae on chickpea, soybean, and maize diets. *J. appl. Ent.*, **123**: 335-339.

SINGH, A.K. AND REMBOLD, H., 1988. Developmental value of chickpea, *Cicer arietinum*, soybean, *Glycine max* and maize, *Zea mays*, flour for *Heliothis armigera* larvae. *J. appl. Ent*., **106**: 286-296.

STEEL, R.G.D. AND TORRIE, J.S., 1980. *Principles and procedures of statistics. A biological approach*. Second Edition, McGraw Hill Book Company Inc., New York, USA.

VANDERZANT, E.S., RICHARDSON, C.D. AND FORT, JR. S.W., 1962. Rearing of bollworm on artificial diet. *J. econ. Ent*., **55**: 140.

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