Original Article

Foraging Behavior of *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae) First Instar Larvae on Selected Cotton Varieties

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Abstract The movement, survival, and weight gain of Helicoverpa armigera Hübner (Lepidoptera: Noctuidae) first instar larvae were studied on CB9, CB10 and SR05 cotton varieties under field conditions. The neonate H. armigera were released on the cotton varieties at the squaring stage of the plants and, after a period of 72 hours, the survival, weight gain, and final location of the larvae were observed. While the different cotton varieties had no effect on the survival and weight gain of the larvae, the release locations on the cotton varieties had a significant influence on the larval survival and weight gain. The larvae fed small squares of the cotton varieties were significantly heavier and showed a higher mortality than the larvae fed leaflets and mature leaves. For the cotton varieties in this study, the larvae released on leaflets showed a significantly higher rate of recovery compared to the larvae released on mature leaves and squares. This study also found that the larvae on leaflets did not move up or downward unlike the larvae on mature leaves and squares. This information on the foraging behavior of larvae on cotton varieties will assist researchers to interpret field data and thereby help with the development of pest management decisions.

Keywords: Cotton variety, Helicoverpa armigera, larval movement

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Introduction

Cotton is a highly valued cash crop in Bangladesh, where the widely cultivated varieties include CB9, CB10, and SR05. While CB9 is a long height (115.0 cm), bushy, and hairy variety with trichomes 193/cm², CB10 is a short height (88.3 cm) and smooth variety, and SR05 is a well ventilated and slightly hairy variety with trichomes 106/cm² (Amin et al., 2008; Amin et al., 2011). Every year a number of sucking and chewing insect species attack these cotton varieties and cause significant crop loss. Among these insect species, the polyphagous insect *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae) is one of the most significant cotton pests in Bangladesh (Amin et al., 2008). Their larvae have been reported to feed on over 130 plant species worldwide (Matthews, 1999), initially eating the tender leaves close to where they hatch and then moving upwards to the reproductive organs (Zalucki et al., 1986; Perkins et al., 2009).

This pest has already developed a resistance to a range of insecticides (Yang et al., 2008), and some insecticides even kill predator and pollinator species (Boyd and Boethel, 1998; Sclar et al., 1998; Moser and Obrycki, 2009). Meantime, resistant varieties have been developed that are safe from pollution and environment friendly, and use cost-cutting methods in integrated pest management programs (Taylor et al., 2001; Nault et al., 2004). The host plant characteristics, such as the waxes, trichomes, cuticle thickness, and toxic allelochemicals, are also known to affect the larval foraging, feeding, growth, and development (Zalucki et al., 2002).

Caterpillars can spend more time for processing a feeding site on leaves with a heavy layer of wax or many trichomes, as they have to remove these obstacles to access more nutritious layers. Furthermore, thick blooms of wax can gum up the larval mouthparts, which then need to be cleaned before the larvae can continue feeding. This pre-processing and preening increases the

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amount of time spent at risk of predation. The surface of the leaves also affects the way the larvae deposit silk on the plant (Varela and Bernays, 1988). The larvae of *H. armigera* deposit silk continuously as they walk across the plant and before they feed (Sorensen et al., 2006). This then affects the larval adherence and locomotion across the leaf surface, as the legs of the lavae can become tangled with the silk threads (Varela and Bernays, 1988), impeding locomotion, and can be hampered by wax crystals (Eigenbrode and Shelton, 1992) or trichomes (Zalucki et al., 2002).

Selecting the proper variety and protecting the crops from pests and diseases are important prerequisites for a higher yield and quality of cotton. Therefore, this study investigated the foraging behaviour of *H. armigera* first instar larvae on the morphologically different cotton varieties of CB9, CB10, and SR05. The resulting data on the foraging behavior of *H. armigera* first instar larvae on these varieties provides basic information on the most vulnerable stage of *H. armigera* with a view to providing information to cotton growers and researchers for proper management of this pest.

Materials and Methods

The study was conducted at the Regional Cotton Research Station, Dinajpur, Bangladesh from August to December 2012.

Collection and rearing of insects

The adult male and female *H. armigera* moths were collected from the Cotton Research Station, Dinajpur. The moths were kept in pairs in Petri dishes $(9.0 \times 1.5 \text{ cm})$ for mating. Every morning, a fresh cotton flower was supplied to the Petri dishes as food for the moths. The Petri dishes were continuously examined to observe mating. After the completion of mating, the male moth was removed from the Petri dish and the female kept in a jar $(26.5 \times 13.5 \text{ cm})$ for oviposition. Cotton leaves and flowers were supplied to the jar. Every morning the jars were cleaned and monitored to observe the egg mass and hatching of eggs. The neonate larvae were used for the experiments.

Observation of foraging behavior

To observe the foraging behavior of the *H. armigera* first instar larvae, the CB9, CB10, and SR05 cotton varieties were grown in earthen pots. The foraging behavior of the larvae was repeated twelve times for each variety using 40-50-day-old-plants (early squaring stage). To test the first instar larval growth and survival, and the feeding locations on the cotton varieties, 10 0-4-h-old larvae were inoculated onto each plant in one of three starting positions: on the upper surface of the top immature leaf or on a mature leaf on the first fruiting branch or square of the plant. For each experiment, 40 larvae were weighed individually to obtain an initial average weight. After 72 h, each plant was carefully searched to recover the live larvae, note their positions, and measure their weights.

Results

Table 1 shows that a total of 360 neonates were placed on each cotton cultivar. After 72 h, a total of 126, 121, and 134 live larvae were recovered from the CB9, CB10, and SR05 variety, respectively. Thus, the overall survival rates for these varieties were, 35.0, 33.6, and 40.3%, respectively, which showed no significant difference (χ^2 =1.0, *p*=0.61). The larval weight gains on the CB9, CB10, and SR05 varieties were 159.3×10⁻⁵,

 Table 1. Overall fate of *Helicoverpa armigera* early instar larvae recorded after 72 h on CB9, CB10, and SR05 cotton varieties

Variety	Total number released	Total number recovered	Survival rate (%)	Weight (mean \pm SE 10 ⁻⁵ g)
CB9	360	126	35.0	159.3±47.8
CB10	360	121	33.6	144.8 ± 37.5
SR05	360	134	40.3	176.5 ± 45.7

 Table 2. Effect of release site (leaflet, mature leaf, or small square) on number and location of first instar *Helicoverpa armigera* larvae recorded after 72 h exposure on CB9, CB10, and SR05 cotton varieties

Variety	Release Site	Total no. released	Number and locations of larvae collected after 72 h				Total no. and rate
			Folded leaflet	Unfolded leaflet	Mature leaf	Small square	(%) recovered
CB9	Leaflet	120	0	8	36	19	63 (52.5%)
	Mature leaf	120	0	7	29	14	50 (41.7%)
	Small square	120	1	3	3	7	14 (11.7%)
CB10	Leaflet	120	0	10	36	13	59 (49.2%)
	Mature leaf	120	0	4	31	14	49 (40.8%)
	Small square	120	0	3	6	4	13 (10.8%)
SR05	Leaflet	120	0	10	42	21	73 (60.8%)
	Mature leaf	120	0	7	32	15	54 (45.0%)
	Small square	120	0	4	6	9	19 (15.8%)

A leaflet is the upper part of a terminal leaf; mature leaf on the first fruiting branch, and square on the first fruiting branch.

Table 3. Mean weight (10^{-5} g) of *Helicoverpa armigera* larvae recovered after 72 h from CB9, CB10, and SR05 cotton cultivars based on initial location (leaflet, mature leaf, or small square)

Cotton variety	Release site	Total no. released	Total no. recovered	Weight (mean SE 10 ⁻⁵ g)
CB9	Leaflet	120	20	159.5±41.9
	Mature leaf	120	65	136.8 ± 60.5
	Small square	120	41	181.6±40.9
CB10	Leaflet	120	20	$137.8 {\pm} 26.6$
	Mature leaf	120	62	123.2 ± 49.9
	Small square	120	39	173.4±35.9
SR05	Leaflet	120	27	172.0 ± 38.8
	Mature leaf	120	64	147.4 ± 31.2
	Small square	120	43	210.1±45.2

144.8×10⁻⁵, and 176.5×10⁻⁵ g, respectively, which also showed no significant difference (χ^2 =3.2, *p*=0.20).

Table 2 shows that the larvae were more likely to be found on a leaflet than on a mature leaf or small square. On the CB9 variety, 52.5, 41.7, and 11.7% of the larvae were found on a leaflet, mature leaf, and square, respectively, indicating a significant difference (χ^2 =25.3, p<0.001). On the CB10 variety, the larval recovery rates on a leaflet, mature leaf, and square were 49.2, 40.8, and 10.8%, respectively, indicating a significant difference (χ^2 =23.8, p<0.001). Finally, the larval recovery from the SR05 variety showed a higher larval population on the leaflets (60.8%), followed by the mature leaves (45.5%), and small squares (15.8), plus the start location was found to have a significant effect on the rate of recovery (χ^2 =25.9, p<0.01).

For the CB9 variety, the release location was found to have a significant effect on the weight gain ($\chi^2=6.3$, p<0.05), where the larvae released on the small squares were heavier (173.6×10⁻⁵ g) than those released on the leaflets (159.5×10⁻⁵ g) and mature leaves (144.8×10⁻⁵ g). The larval release location on the CB10 variety also had a significant effect on the weight gain ($\chi^2=9.1$, p<0.01), where the larvae released on the small squares were heavier (173.6×10⁻⁵ g) than those released on the leaflets (159.5×10⁻⁵ g) and mature leaves (144.8×10⁻⁵ g). Similarly, the weight gain was significantly affected by the larval release location on the SR05 variety ($\chi^2=11.4$, p<0.01), where the larvae released on the small squares were heavier (173.6×10⁻⁵ g) than those released on the small squares (144.8×10⁻⁵ g).

Discussion

The manipulation of plant attributes through breeding and culturing practices has the potential to protect crops from economic damage due to feeding herbivores by altering their feeding site selection. Various researchers have already built descriptions of the foraging behavior and movement of *H. armigera* larvae on plants (Zalucki et al., 2002; Johnson and Zalucki, 2005; Johnson and Zalucki, 2007; Johnson et al., 2007). Newly hatched *H. armigera* initially rest under a leaf for about 2 h. The larvae then enter an active phase, generally moving in an upward direction and feeding intermittently on small-sized meals. After several hours, the larvae feed more constantly and move around. Not all the larvae move from the plant component where they hatch, and the distances moved by the larvae vary considerably.

Young Lepidoptera larvae generally display a replicable behavior moving up their host plant (Perkins et al., 2008). The nitrogen and water contents of food are important determinants of young caterpillar performance (Zalucki et al., 2002). Thus, since the leaves in the lower/ older plant regions contain the least proportion of nitrogen and seem to be of least nutritive value, the larvae move up the plant.

The leaflet hardness varied considerably among the cotton varieties used in this study. As a result, hardness was discounted as a macro-level factor influencing the movement of the first instars. However, the hardness of the plant components likely influenced the movement on a local (micro) scale, where the hard plant parts were 'ignored' as a potential food source and the larva moved on until softer plant tissue was encountered. Thus, the movement of young caterpillars on plants is assumed to be a variable between Lepidoptera species and between host plants.

The polyphagous insect considered in the current study is one of the most damaging pests for field crops in Bangladesh, and chemical insecticides have traditionally been used to control this pest. Yet this management technology disrupts the environment, and the renewed emphasis on sustainable, environment-friendly crop protection practices has highlighted the need to develop alternative pest management strategies. Accordingly, the present study generated important data that can hopefully be used for the development of integrated management of *H. armigera* in the cotton fields of Bangladesh.

References

- Amin MR, Ahad MA, Hossain MH, Hossain SMA, Tithi DA (2008) Characteristics of some cotton varieties in relation to seasonal abundance of pests, predators and their impact on yield and quality. J Agrofor Environ 2: 67-70.
- Amin MR, Tithi DA, Kwon YJ (2011) Characteristics of three cotton varieties and their impact on feeding and growth of cotton armyworm. Entomol Res 41: 151-156.
- Boyd ML, Boethel DJ (1998) Residual toxicity of selected insecticides to heteropteran predaceous species (Heteroptera: Lygaeidae, Nabidae, Pentatomidae) on soybean. Environ Entomol 27: 154-160.
- Eigenbrode SD, Shelton AM (1992) Survival and behavior of *Plutella xylostella* larvae on cabbages with leaf waxes altered by treatment with S-ethyl dipropylthiocarbamate. Entomol Exp Appl 62: 139-145.

- Johnson ML, Armitage S, Scholz BCG, Merritt DJ, Cribb BW, Zalucki MP (2007) Predator presence moves *Helicoverpa armigera* larvae to distraction. Insect Behav 20: 1-18.
- Johnson ML, Zalucki MP (2005) Foraging behaviour of *Helicoverpa* armigera first instar larvae on crop plants of different developmental stages. J Appl Entomol 129: 239-245.
- Johnson ML, Zalucki MP (2007) Feeding and foraging behaviour of a generalist caterpillars: are third instars just bigger versions of firsts? Bull Entomol Res 97: 81-88.
- Matthews M (1999) Heliothine moths of Australia: A Guide to Pest Bollworms and Related Noctuid Groups. CSIRO Publishing, Collingwood, Victoria, Australia.
- Moser SE, Obrycki JJ (2009) Non-target effects of neonicotinoid seed treatments; mortality of coccinellid larvae related to zoophytophagy. Biol Control 51: 487-492.
- Nault BA, Taylor AG, Urwiler M, Rabaey T, Hutchison WD (2004) Neonicotinoid seed treatments for managing potato leafhopper infestations in snap bean. Crop Prot 23: 147-154.
- Perkins LE, Cribb BW, Hanan J, Glaze E, Beveridge C, Zalucki MP (2008) Where to from here? The mechanisms enabling the movement of first instar caterpillars on whole plants using *Helicoverpa armigera* (Hübner). Arthropod Plant Interact 2: 197-207.
- Perkins LE, Cribb BW, Hanan J, Zalucki MP (2009) The role of two plantderived volatiles in the foraging movement of 1st instar *Helicoverpa armigera* (Hübner): time to stop and smell the flowers. Arthropod

Plant Interact 3: 173-179.

- Sclar DC, Gerace D, Cranshaw WS (1998) Observations of population increases and injury by spider mites (Acari: Tetranychidae) on ornamental plants treated with Imidacloprid. J Econ Entomol 91: 250-255.
- Sorensen GS, Cribb BW, Merritt D, Johnson ML, Zalucki MP (2006) Structure and ultrastructure of the silk glands and spinneret of *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae). Arthropod Struct Dev 35: 3-13.
- Taylor AG, Eckenrode CJ, Straub RW (2001) Seed coating technologies and treatments for onions: challenges and progress. Hort Sci 36: 199-205.
- Varela LG, Bernays EA (1988) Behavior of newly hatched potato tuber moth larvae, *Phthorimaea operculella* Zell. (Lepidoptera: Gelechiidae), in relation to their host plants. J Insect Behav 1: 261-275.
- Yang Y, Johnson ML, Zalucki MP (2008) Possible effect of genetically modified cotton on foraging habits of early instar *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) larvae. Aust J Entomol 47: 137-141.
- Zalucki MP, Clarke AR, Malcolm SB (2002) Ecology and behavior of first instar larval Lepidoptera. Annu Rev Entomol 47: 361-393.
- Zalucki MP, Daglish G, Firempong S, Twine P (1986) The biology and ecology of *Heliothis armigera* (Hübner) and *Heliothis punctigera* Wallengren (Lepidoptera: Noctuidae) in Australia-what do we know? Aust J Zool 34: 779-814.