

## Natural Mortality of *Helicoverpa armigera* (Hübner) Eggs in the Cotton Ecosystem

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### ABSTRACT

Field studies were conducted during the cotton crop seasons in 2003 and 2004 at CCS Haryana Agricultural University, Hisar, to identify mortality factors in the egg stage of *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) in the insecticide-free cotton fields to gain an insight into the role of these factors in causing egg specific mortality. Eggs were collected from cotton fields on different dates during September-November and brought to the laboratory for determining egg viability and level of parasitism. Similarly, plant parts bearing eggs in the field were tagged on different dates and observed daily to assess egg loss. The correlation of egg loss with various abiotic factors was determined. The mean viability of field collected eggs was 75% and 89% during 2003 and 2004, respectively (pooled mean egg viability of 82%). It was 51.34-66.55 (Mean: 59.72) % in the case of eggs deposited in the field by the laboratory-reared females. Eggs were parasitized by *Trichogramma chilonis* Ishii during September-October and mean parasitization was 25 and only 1-2% during 2003 and 2004, respectively. There was a positive correlation of % egg loss with wind velocity and rainfall as evident for multiple correlation coefficient values, i.e. 0.92 and 0.60 for 2003 and 2004, respectively. A partial ecological life table constructed for egg stage indicated that 23-44% mortality occurs at this stage. Important factors responsible for egg mortality/loss were identified as *T. chilonis*, inviability, rain, wind and unexplained mortality.

**Keywords:** Cotton, Egg mortality, *Helicoverpa armigera*, Lepidoptera, Noctuidae, Parasitization, *Trichogramma chilonis*.

### INTRODUCTION

The increase and decrease of populations of living organisms is related to the capacity of the individuals to survive and reproduce. Members of a species increase as long as intrinsic and extrinsic factors favour that species. A species encounters various abiotic and biotic stresses at different periods. Successful survival is governed by the intensity of the stresses, as the future build-up of population depends on the proportion of individuals surviving through various stresses at different stages. Among insects, certain species attain the status of pest when conditions become favourable. The

noctuid, *Helicoverpa armigera* (Hübner) (Lepidoptera), is a major pest of many economically important crops in India. This pest is active throughout the year on one or other crops. A wide host range (Reed and Pawar, 1982; Fitt, 1989; Zalucki *et al.*, 2002), high fecundity (Reed, 1965), a capacity to migrate (Farrow and Daly, 1987) and the ability to develop high resistance to insecticides (McCaffery *et al.*, 1986) have enabled this insect to attain key pest status among various major crop pests. In Haryana, it has gradually elevated its position to become the major pest of cotton during the last two decades ever since it was first recorded on this crop in 1978 (Jayaswal and Singh, 1979). Some 14-56% of

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the cotton crop is damaged by *H. armigera* (Kaushik et al., 1969; Manjunath et al., 1989; Jayaraj, 1990).

Predicting the role of natural mortality in population suppression and regulation and identifying the key mortality agents responsible for the biotic component of natural mortality provide the basic ingredients for developing biologically intensive strategies for managing bollworms without dependence on insecticides (Sansone and Smith, 2001). The egg stage is very crucial in the life cycle of *H. armigera* as high mortality is expected in this stage. Kyi et al. (1991) reported up to 88% mortality of *H. armigera* eggs mostly within the first 3 days of oviposition by females. Similarly, Sansone and Smith (2001) reported 71-95% mortality at the egg and first larval stages.

Early season application of broad spectrum insecticides, often targeted against jassid (*Amrasca biguttulla biguttulla* Ishida), generally kills the natural enemies of bollworms and reduces the impact of natural enemies on subsequent bollworm densities later in the season (King and Coleman, 1989). If the role of specific natural enemies in *H. armigera* suppression can be identified and measured in insecticide-free cotton, then an 'insecticide predisposition hypothesis' to explain consistent bollworm outbreaks can be better addressed. Hence, the present study was planned to gain an insight into the role of various mortality factors in the stage specific mortality of *H. armigera* eggs so that these can be better exploited for the management of this notorious pest.

## MATERIALS AND METHODS

The experiment was conducted in the laboratory as well as at the Research Farm of the Department of Entomology, Chaudhary Charan Singh Haryana Agricultural University at Hisar (29° 10' N, 75° 46' E, 215.2 m m.s.l.) in Haryana (India) during the cotton crop seasons of 2003 and 2004. The cotton variety H-1098 (available from Cotton Section, Department of Plant Breeding, CCS Haryana Agricultural University, Hisar, Haryana, India) in 2003 and 2004 was sown in a 25×50 m plot in the first

week of May. All the recommended agronomic practices were followed except for the spraying of insecticides (Anonymous, 2004). The plant to plant and row to row spacing was 30 and 67.5 cm, respectively. Cotton flowering started in July end to early August.

Survival of the *H. armigera* eggs was assessed using two methods. In the first method, leaves bearing *H. armigera* eggs were detached from the cotton plants in insecticide-free fields at weekly intervals during September-October and brought to the laboratory. A simple random sampling method was followed for the collection of eggs. Before sampling, the field was divided into four quarters, each quarter representing one replication, with a total of four replications. From sampled leaves, the egg bearing portion was cut and placed in individual glass vials (5 cm×1.25 cm) to record hatching and parasitization, if any. Depending upon the availability of eggs, a weekly sample of 25 to 150 eggs was examined. Additionally, eggs were collected from farmers' fields (village Kaimari and Ludas) near Hisar and were observed for hatching and parasitization as described above.

In the second method, three pairs of 2-3 day-old laboratory reared *H. armigera* male and female moths were confined in each of three muslin cages (30 cm × 20 cm) over cotton branches in the field for oviposition. Each cage represented one replication with a total of three replications. A cage was supported by an iron frame which was tied to a stick fixed in the ground. After two days, the cages were removed and the plants were observed to identify the number of eggs laid. After four days (when the eggs reached black head stage) the eggs were collected along with the substrate in the glass vials and kept under observation to record hatching and parasitization, if any. A record of the eggs found missing from the plants was kept. Weekly data on wind velocity and average rainfall were recorded at the Meteorological observatory located at about 0.75 km from the experiment site. The relationship of egg loss with respect to rainfall and wind velocity was worked out. Various factors responsible for egg loss/mortality were identified. For example, eggs that did not hatch

for more than 4 or 5 days and turned charcoal black in color were observed for any possible parasitism and those which remained creamy white but did not hatch without showing any external sign of parasitization or predation were considered inviable. The disappearance of eggs was due to unexplained mortality. A partial ecological life table was constructed in order to understand the role each factor played in causing egg stage mortality (Southwood, 1978). The significance of difference in egg viability and parasitization in 2003 and 2004 was analyzed by Student's t-test (Panse and Sukhatme, 1995).

## RESULTS AND DISCUSSION

### Egg Viability

#### Field Collected Eggs Naturally Laid

In 2003, egg hatchability was 71% during September while it was 60.31 to 86.58 during October with a mean hatchability of 75.12% (Table 1). Similarly, during 2004, egg hatchability ranged from 85.21-85.87, 87.03-96.00 and 100% during September, October and the first week of November, respectively, with a mean hatchability of 88.66% (Table 1). It was significantly higher during 2004 than 2003. The pooled mean of two years indicated that

82.34% of eggs were viable.

#### Eggs Deposited in Field by Laboratory Reared Females

In 2003, egg hatchability was 18.34 to 73.04% during September and 31.45 to 77.77% during October with a mean hatchability of 51.34% (Table 2). Similarly, during 2004, it ranged from 35.63 to 79.62% in September and 32.60 to 89.58% in October with a mean hatchability of 66.55%. It was significantly higher during 2004 than during 2003. Pooled mean egg viability for the two years was 59.72%.

It could be inferred from the data that egg viability was significantly higher during 2004 compared to that during 2003 both in the field collected eggs as well as in those deposited by laboratory reared females. Further, it was considerably higher in the field collected eggs than in those laid by laboratory reared females as a greater proportion of the latter was found parasitized (Discussed elsewhere). The higher egg viability and consequent low parasitization during 2004 may be attributed to low parasitoid activity during September-October consequent to no or low host eggs availability during June to August. The temperature and relative humidity during these months fluctuated between 35-42°C and 25-59%, respectively (data not presented). Further, there was no rain

**Table 1.** Hatching and parasitization in eggs of *H. armigera* collected from cotton during 2003 and 2004.

Month	Week	Number of eggs collected		Egg hatching (%)		Parasitization (%)		
		2003	2004	2003	2004	2003	2004	
September	IIIrd	--	142	--	85.21	--	0.00	<i>Trichogramma chilonis</i> Ishii
	IVth	150	92	71.33	85.87	28.66	0.00	"
October	Ist	63	54	60.31	87.03	39.69	0.00	"
	IInd	82	50	86.58	92.00	13.41	6.00	"
	IIIrd	91	50	81.32	96.00	18.68	4.00	"
November	IVth	--	28	--	89.28	--	3.57	"
	Ist	--	25	--	100.00	--	0.00	--
Total/ Mean *		386	441	75.12*	88.66*	24.87*	1.36*	
					0.01		0.02	

t ( $p \leq 0.05$ )

**Table 2.** Hatching, parasitization and loss of eggs laid by laboratory reared *H. armigera* females during 2003 and 2004.

Month	Week	Number of eggs collected		Egg hatching (%)		Parasitization (%)			Egg loss (%)	
		2003	2004	2003	2004	2003	2004		2003	2004
September	IInd	115	87	73.04	35.63	19.13	0.00	<i>Trichogramma chilonis</i> Ishii	4.34	59.77
	IVth	109	108	18.34	79.62	9.17	0.00	"	72.47	15.74
October	IInd	124	138	31.45	32.60	62.90	1.45	"	5.56	67.50
	IIIrd	--	144	--	89.58	--	6.94	"	--	2.08
	IVth	135	115	77.77	89.56	12.59	2.61	"	5.93	4.35
Total/Mean*		483	592	51.34*	66.55*	26.29*	2.53*		20.49*	28.04*
				0.13		0.05			0.28	

t (p≤ 0.05)

even for a single day during July which led to drought like conditions. Consequently, no host or parasitoid activity was observed during these months in 2004 and the parasitoid population build up during the subsequent months was also low. This led to a higher proportion of eggs remaining unparasitized. No information is available on the viability of *H. armigera* eggs on cotton under field conditions. However, egg viability of 56.4 (Singh, 2000) and 62.4-89.0% (Chaudhary and Sharma, 1981) in *H. armigera* eggs was reported on chickpea. Thus, the viability of *H. armigera* eggs was quite high under the cotton ecosystem.

### Egg Parasitization

#### In Field Collected Eggs

During 2003, parasitization in field collected *H. armigera* eggs by *Trichogramma chilonis* Ishii was 29% by the last week of September and 13.41 to 39.69% in October with maximum parasitization in the first week (Table 1). The pooled mean egg parasitization was 24.87%. During 2004, no egg parasitization was observed in September while it varied from 3.57 to 6.00% during October. It was significantly lower during 2004 than during 2003. Pooled mean parasitization for the two years was 12.33%. Earlier, field parasitization of *H. armigera* eggs on cotton by *T. chilonis* ranging from 18.18 to 82.76% was reported by Pascua and Pascua (1995). Suphankasen (1979) recorded 80% egg loss of *H.*

*armigera* due to parasitization by *T. chilonis*.

#### In Eggs Deposited in Field by Laboratory Rereared Females

During 2003, egg parasitization was 9.17 to 19.13% during September and 12.59 to 62.90% during October with mean parasitization of 26.29%. During 2004, no parasitization was observed in September while it ranged from 1.45 to 6.94% during October with mean parasitization of 2.53%. Parasitization in 2004 was significantly lower than that in 2003. Overall mean parasitization for both years was 13.21%. As compared to field collected eggs, parasitization in the eggs deposited by laboratory reared females was somewhat higher in the second week of October, 2003. This is probably due to the availability in the latter case of a larger number of eggs on a single plant for parasitization by *T. chilonis*. Parasitization by *Trichogramma* spp. has been reported to be mostly density dependent (ICRISAT, 1988).

A variable percentage of eggs was found missing during the tow years. Mean egg loss was 20.49% (4.34-72.47%) during 2003 and 28.04% (2.08-64.50%) during 2004.

It was concluded from these studies that egg parasitization of *H. armigera* by *T. chilonis* was highest during the first fortnight in October. It was low during 2004 as compared to 2003. Low parasitization during 2004 was probably due to

unfavorable environmental conditions (high temperature, low rainfall and low relative humidity) prevailing during the season resulting in poor build up of *H. armigera* population.

#### ***H. armigera* Egg Loss in Relation to Weather Parameters**

Both during 2003 and 2004, a direct and positive correlation was observed between % egg loss due to dislodgement from plants and wind velocity ( $r=0.87$  and  $0.70$  for 2003 and 2004, respectively) (Table 3). Similar correlation was observed between egg loss and rainfall ( $r=0.95$  and  $0.62$  for 2003 and 2004, respectively). The multiple correlation coefficient ( $R$ ) for % egg loss and wind velocity and rainfall was  $0.92$  and  $0.60$  for 2003 and 2004, respectively.

During 2003, egg loss as high as  $80.00$  and  $72.47\%$  was observed in eggs laid on 26 and 28 September, respectively. This was ascribed to heavy rains ( $23$  mm) and strong winds ( $8.9$  km hr<sup>-1</sup>) recorded on 29 September.

Similar levels of egg loss was observed during 2004. It was  $59.77\%$  for eggs laid on 9 September which corresponded with high wind velocity ( $6.4$  km hr<sup>-1</sup>) on 9 and 10 September. Similarly, this loss was  $64.5\%$  in eggs laid on 8 October which corresponded with a high wind velocity ( $7.3$  to  $12.6$  km hr<sup>-1</sup>) on 10 and 11 October coupled with  $8$  mm rains on 10 October. Kyi *et al.* (1991) reported egg losses to be  $32$ – $68\%$  within the first three days of oviposition in *H. armigera*, among which wind, rain and aerial predators were the major mortality factors.

#### **Mortality Factors of *H. armigera* Eggs**

In September, 2003,  $1.07\%$  did not hatch while  $20.05$  were parasitized by *Trichogramma chilonis* (Table 4). A greater percentage ( $22.46$ ) was missing from the plants and was included in unexplained

mortality. Similarly in October,  $1.01\%$  did not hatch while  $29.90\%$  were parasitized by *T. chilonis*. A small percentage ( $3.03$ ) of eggs was missing from the plant surface.

In September 2004,  $10.02\%$  did not hatch while no egg was parasitized in this month. However,  $16.08\%$  eggs were missing from the plants. In October,  $3.10\%$  eggs failed to hatch while  $3.62\%$  were parasitized by *T. chilonis*. About  $16.75\%$  eggs were found to be missing from the plants.

The overall trend of mortality due to different factors during both the years indicated that the removal of eggs due to various reasons (wind/rain/unexplained reasons), parasitization by *T. chilonis* and inviability were the most important egg mortality factors which accounted for  $10.42$ – $19.05$ ,  $9.34$ – $15.73$  and  $2.14$ – $5.85\%$  egg mortality, respectively. The total egg mortality due to various reasons varied from  $23.48$  to  $43.58\%$ . The various biotic mortality factors that normally existed in the cotton field were *Trichogramma chilonis*, *Chrysoperla carnea* and Coccinellids; the latter two may be responsible for the removal of a proportion of eggs which were included in unexplained mortality.

Zalucki *et al.* (2002) stated that in Lepidoptera generally about  $41\%$  eggs are lost before hatching due to various reasons. Similarly, Sansone and Smith (2001) also reported  $71$ – $95\%$  mortality of *Helicoverpa zea* at the egg and first-instar larval stages in short season cotton in Texas. Among the various mortality factors, they reported unexplained mortality to be the leading cause, while parasitization by *Trichogramma* spp. accounted for only  $3.4\%$ . In the present studies, even during the period when there were no rains and also wind velocity was low, a small percentage of eggs ( $<6\%$ ) were found missing from the plants. It was difficult to assign their losses to a particular agent. Nevertheless, predation or their dislodgement due to plant or unexplained mortality could be one of the reasons. Kyi *et al.* (1991) attributed  $5$ – $6\%$  egg loss in *H. armigera* to changing leaf orientation.



**Table 3.** Correlation of % egg loss of *H. armigera* with wind velocity and rainfall during cotton crop seasons 2003 and 2004.

Date of egg laying	% egg loss (up to 4 days after egg laying)	Wind velocity (km/h) (Days after egg laying)					Rainfall (mm) (Days after egg laying)					Total	
		0	1	2	3	4	Mean	0	1	2	3		4
14 Sept., 2003	4.34	3.5	2.0	4.9	3.5	4.2	3.62	0.0	0.0	2.2	0.0	0.0	2.2
19 Sept., 2003	28.00	3.7	3.0	2.8	1.8	5.5	3.36	0.0	0.0	0.0	0.0	0.0	0.0
26 Sept., 2003	80.00	8.9	3.5	4.5	8.9	4.3	6.02	0.0	0.0	0.0	23.0	0.0	23.0
28 Sept., 2003	72.47	4.5	8.9	4.3	2.2	2.5	4.48	0.0	23.0	0.0	0.0	0.0	23.0
29 Sept., 2003	0.70	2.1	4.3	2.2	2.5	2.4	2.70	0.0	0.0	0.0	0.0	0.0	0.0
06 Oct., 2003	5.46	2.4	4.2	1.9	1.2	1.4	2.22	0.0	0.0	0.0	0.0	0.0	0.0
14 Oct., 2003	5.65	2.7	2.6	1.7	1.8	1.8	2.12	0.0	0.0	0.0	0.0	0.0	0.0
28 Oct., 2003	5.93	1.4	1.5	1.1	0.8	0.8	1.12	0.0	0.0	0.0	0.0	0.0	0.0
Corr. Coeff. (r)							0.87						0.95
Multiple Corr. Coeff. (R)													0.92
09 Sept., 2004	59.77	6.4	6.4	4.7	5.1	3.3	5.18	0.0	0.0	0.0	0.0	0.0	0.0
26 Sept., 2004	15.74	2.2	2.6	2.5	3.3	6.0	3.32	0.0	0.0	0.0	0.0	0.0	3.0
27 Sept., 2004	23.53	2.6	2.5	3.3	6.0	2.9	3.46	0.0	0.0	0.0	3.0	0.0	3.0
04 Sept., 2004	16.00	4.2	1.9	1.3	1.0	1.7	2.02	0.0	0.0	8.0	0.0	0.0	0.0
08 Oct., 2004	64.50	1.7	3.6	7.3	12.6	3.6	5.76	0.0	0.0	0.0	0.0	0.0	8.0
11 Oct., 2004	4.00	12.6	3.6	3.7	1.8	2.0	4.74	0.0	0.0	0.0	0.0	0.0	0.0
18 Oct., 2004	2.08	1.5	4.3	3.2	6.0	3.0	3.66	0.0	0.0	0.0	0.0	0.0	0.0
27 Oct., 2004	10.71	2.8	2.7	6.3	2.5	1.0	3.06	0.0	0.0	0.0	0.0	0.0	0.0
29 Oct., 2004	4.35	6.3	2.5	1.0	1.3	1.3	2.48	0.0	0.0	0.0	0.0	0.0	0.0
Corr. Coeff. (r)							0.70						0.62
Multiple Corr. Coeff. (R)													0.60

**Table 4.** Mortality factors for *H. armigera* eggs as summarized in the form of partial ecological life table.

Month	No. of eggs Observed (lx)			Factors responsible for dx (dxF)	No. dying during x (dx)			qx (%) <sup>a</sup>		
	2003	2004	Total		2003	2004	Mean	2003	2004	Mean
September	374	429	803	Inviability	4	43	47	1.07	10.02	5.85
				Parasitization	75	0	75	20.05	0.00	9.34
				Unexplained mortality	84	69	153	22.46	16.08	19.05
				Total	163	112	275	43.58	26.10	34.25
October	495	579	1074	Inviability	5	18	23	1.01	3.10	2.14
				Parasitization	148	21	169	29.90	3.62	15.73
				Unexplained mortality	15	97	112	3.03	16.75	10.42
				Total	168	136	304	33.94	23.48	28.30

$$^a qx = dx/lx * 100$$

The activity of *T. chilonis* against *H. armigera* populations in the cotton growing area of south-western Haryana (i.e. Hisar and adjoining districts) appears to depend largely upon the prevailing weather conditions. This is strongly supported by the high rate of parasitization observed in 2003 which received 431.99 mm rains than that in 2004 receiving only 192.3 mm rainfall from July to October. Further, there were no rains during July 2004 leading to drought like conditions. Consequently, a higher *H. armigera* population was observed during 2003 as compared to 2004 (Kumar, 2005) thus ensuring host availability for *T. chilonis*. Hence, in the current insect-pest management schemes, age-specific intrageneration mortality is an important parameter for predicting the fate of the pest population i.e. the proportion of eggs that will survive to the damaging larval stage. Cotton pest management is dependent upon insecticide application decisions that are initiated when scouting estimates of egg and early instar densities are approaching Economic Threshold Levels (ETLs). Bollworm eggs present the easiest (and possibly the most accurate) life stage for census (Sansone and Smith, 2001). However, insecticide application decisions should not be solely based on egg densities because a high level of mortality is expected to occur at this stage. Thus, a delayed application of insecticides until all the mortality factors have affected the early stages of bollworms can form an important component of

Integrated Pest Management. Moreover, if insecticide applications are to be made, selective insecticides such as endosulfan should be used to conserve the natural enemies at the early fruiting phase of the crop.

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مرگ و میر طبیعی تخمهای *Helicoverpa armigera* (Hüber) در اکوسیستم پنبه  
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چکیده

به منظور درک نقش عوامل مرگ و میر مختص مرحله تخم، مطالعاتی در دانشگاه کشاورزی هاریانا طی سالهای ۲۰۰۳، ۲۰۰۴ برای تشخیص عوامل مرگ و میر تخم های *Helicoverpa armigera* در مزارع عاری از حشره کش انجام شد. برای تعیین قدرت بقا و میزان پارازیتسم، تخمها در زمانهای مختلف طی ماههای شهریور تا آبان ماه از مزارع جمع آوری و به آزمایشگاه منتقل شدند. همچنین بخشهایی از گیاهان حاوی تخم در زمانهای مختلف محصور شده و روزانه برای ارزیابی میزان اتلاف تخمها بررسی می شدند. همبستگی بین اتلاف تخم و عوامل غیر زنده متعددی تعیین شد. میانگین بقا تخمهای جمع آوری شده طی سالهای ۲۰۰۳ و ۲۰۰۴ بترتیب ۷۵ و ۸۹ درصد بود (با میانگین کل ۸۲ درصد). این مقدار در مورد تخمهای گذاشته شده بوسیله ماده‌ای حاصل از پرورش آزمایشگاهی بین ۵۱/۳۴ و ۶۶/۵۵ درصد (با میانگین ۵۹/۷۲ درصد) بود. تخمها طی ماههای شهریور تا آبان ماه به وسیله زنبور *Trichogramma chilonis* Ishii پارازیت شده و میانگین پارازیتسم ۲۵ درصد و در سالهای ۲۰۰۳ و ۲۰۰۴ تنها ۱ تا ۲ درصد بود. همبستگی مثبتی بین میزان اتلاف تخم و سرعت باد و باران وجود داشت، به طوری که مقادیر ضریب همبستگی برای سالهای ۲۰۰۳ و ۲۰۰۴ به ترتیب ۰/۹۲ و ۰/۶۰ بود. جدول زندگی جزئی تهیه شده برای مرحله تخم نشان دهنده ۲۳ تا ۴۴ درصد مرگ و میر در این مرحله بود. مهمترین عوامل مرگ و میر تخمها زنبور پارازیت *Trichogramma chilonis*، عدم بقا، باران، باد و عوامل ناشناخته تشخیص داده شدند.