# IMPLICATIONS FOR BT COTTON'S REFUGEE STRATEGY AND FITNESS COSTS OF HELICOVERPA ARMIGERA RESISTANCE

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## Abstract

In order to get the H. armigera larva, many trips were made to the Bt cotton fields in the Karjan area of the Vadodara district of Gujarat. The study was carried out every week throughout the 2019-2020 growing season to gather the population of Bt resistant H. armigera larvae from 105 cotton fields. Five species of Lepidoptera, four species of Hemiptera, and one species of Spotted Wing Drosophila were among the other insect fauna collected alongside 71 Bt-resistant H. armigera larvae and the various plant parts (tender leaves, older leaves, squares, flowers, and bolls) on which they feed during the course of the survey.

***Keywords:*** *Bt Cotton's, Refugee Strategy, Fitness Costs, Helicoverpa Armigera.*

## 1. Introduction

The cotton plant (Genus: Gossypium), sometimes known as "white gold" or "the king of fibres," thrives in India's varied agro-climatic zones. The textile sector in India relies on cotton more than any other single crop, and cotton accounts for almost 65% of the country's overall raw material requirements. India has the most cotton land in the world (12,000 hectares, or 33% of the total), yet its output ranks only second behind China's (5.45 million metric tonnes). It's an important part of the Indian economy since it's the primary raw material used in the textile industry, which employs millions of people. Since cotton is so ubiquitous in our daily lives, it is sometimes called "the mother of civilization" due to its central role in our economics and culture. About 5.8 million people worldwide make their living as cotton farmers, while another 40–50 million people work in industries directly connected to cotton farming, such as cotton processing and trading. Cotton is grown on over 126.07 million hectares of land in India, making it the biggest cotton-growing region in the world. Gossypium arboreum, G. herbaceum, G. barbadense, and G. hirsutum (Asian, Egyptian, and American Upland cotton, respectively) are all grown in India. In India, 88% of hybrid cotton is Gossypium hirsutum, and all contemporary Bt cotton hybrids are G.hirsutum.[1-2]

According to the global cotton situation, India is responsible for 37.56% of global cotton acreage and 24.26% of global cotton output. China is the world's second-largest cotton producer, accounting for 9.97% of cotton land and 22.41% of cotton output. Approximately 74% of global cotton is produced in India, China, the United States, and Brazil. Domestic consumption in China is anticipated to be highest, at 365 lakh bales, or 30.84 percent of world consumption, with India as the second-largest consumer, at 245 lakh bales, or 20.7 percent of global consumption.[3]

According to the CAB study, India has the most cotton acreage in the world at 126.07 lakh hectares, or almost 37% of the total global area of 336.3 lakh hectares. About 62% of India's Cotton is grown in rainfed regions, while the remaining 38% is grown on irrigated grounds. When compared to the United States (955 kgs/ha) and China (1764 Kgs/ha), India's production levels are much lower. It is predicted that India will produce 454.43 kg/ha in 2018-2019.[4]

About 69.33% of India's total cotton producing land and 62.61% of the country's total cotton output may be attributed to the states of Maharashtra, Gujarat, and Telangana. When it comes to land area (26.59lakh hectares) and output (87.00lakh bales of 170 kg), Gujarat is second only to Punjab. Cotton production in Gujarat is more productive than the rest of the country (556.22 kg/ha vs. 454.43 kg/ha).In 2018–19, the nation produced 337 lakh bales (1 bale=170 kg) of cotton over an area of roughly 126.07 lakh hectares, making it the world’s greatest cotton producer.[5]

Insect pest damage prevents India from increasing cotton output to satisfactory levels. Cotton production is very sensitive to environmental factors, insect pests, plant diseases, and agricultural techniques. More than 160 different insect pests cause harm to cotton from the time it emerges until it is harvested. Sap-feeding insects like aphids (Aphis gossypii Glover), jassids (Amrasca biguttula biguttula Ishida), whiteflies (Bemisia tabaci Gennadius), and thrips (Thrips tabaci Lindeman) cause significant damage to cotton crops because they can multiply quickly and in large numbers. There are many species of bollworms that may cause harm to cotton crops during the reproductive phase, including the American bollworm (Helicoverpa armigera) (Hubner), the spotted bollworm (Earias vittella) (Fabricius), the spiny bollworm (Earias insulana) (Biosdual), and the pink bollworm (Pectinophora gossypiella) (Saunders). Pink bollworm infestations caused cotton output losses of 30–80 percent.[6-7]

## 2. Literature review

**Alstad, D. N., & Andow, D. A. (2020)**In order to determine the frequency of Bt-resistance alleles in field populations, a modification of the F1 screen was created to screen F1 progeny derived from single pair mating between field-collected males and laboratory resistant females (designated as F1 screen). However, after treating the F1 progeny derived from more than 260 single pair mating lines with Bt cotton, researchers discovered that there was no clear separation of resistant genotypes from susceptible genotypes based When the body weight of the F1 larvae was 0.7 mg, the association was at its highest.[8]

**Babu, B. G., & Balasubramanian, G. (2019)**The cotton bollworm (Helicoverpa armigera) was the primary focus of an investigation that revealed field-evolved resistance to Cry1Ac in northern China. In a laboratory test, 13 field populations from northern China, where Bt cotton has been planted extensively, were shown to be more resistant to Cry1Ac than two populations from places in northwestern China, where exposure to Bt cotton has been minimal. Resistance to Cry1Ac in northern China is an explicit adaptation caused by exposure to this toxin in Bt cotton, as there was no difference in susceptibility to Bt toxin Cry2Ab between the north and northwest of China.[9]

**Dhaliwal, G. S. & Dhawan, A. K. (2018)**larvae of H. armigera were fed Bt cotton plant parts in a laboratory bioassay to determine the impact of the Cry 1 Ac protein on their survival and development; the results showed that more H. armigera larvae were killed by the leaves of the Bt cotton plants than by the squares of either Bt hybrid. Furthermore, Bt hybrids' leaves and squares had a greater death rate for early larval instars compared to late larval instars. Consistent exposure to Bt cotton plant parts did increase the survival rate of later instar larvae, but it also led to a decrease in larval and pupal weight, an extension of the larval developmental period, malformations in the pupae and adults, a decrease in adult emergence, less fecundity, and low growth and survival indices for both Bt hybrids and their non-Bt counterparts and the check hybrid.[10]

**Bajya, D. R., & Monga, D. (2017)**Researchers found that compared to susceptible larvae, resistant strains took significantly longer to complete development on untreated diet. This finding is consistent with the hypothesis that resistant and susceptible insects develop at different rates. Semi-dominant inheritance may explain why the resistant parent and F1 offspring fared better than susceptible strains given the altered diets. Heterozygotes have an advantage in terms of fitness compared to those who have the sensitive gene, suggesting that resistance has evolved quickly.[11]

**Deevey Jr, E. S. (2016)**the relative fitness of B. thuringiensis (Bt)-susceptible and -resistant colonies was examined by measuring the development time and survival of the Indian meal moth, Plodia interpunctella (Hübner). A fitness cost was associated with resistance to Bt in certain Bt-resistant colonies, but not others, as measured by larval development time and survival. There were clear disparities in maturation lag and mortality rate amongst groups from different regions. It is unknown if variations in the development of susceptible and resistant moths on Bt-treated vs untreated diets will affect the success of mating between the two groups.[12]

## 3. Methodology

Anand Agricultural University in Anand, Gujarat, is home to the All India Network Project on Vertebrate Pest Management. The current research on the fitness costs of Bt resistance in Helicoverpa armigera (Hübner) in Bt cotton is being conducted.

### 3.1 Study area

The study was conducted in Gujarat's Vadodara district, which is a key cotton-growing region. Central Gujarat, or agroclimatic zone III, includes the Vadodara district. The coordinates for the Vadodara district are about 72 degrees 51 minutes east of the Prime Meridian and 21 degrees 49 minutes north of the Equator.

### 3.2 Screening and isolation of bt resistance in h. armigera

In 2020, the AINPVPM Laboratory at Anand Agricultural University in Anand carried out an experiment to screen for and isolate Bt resistance in H. armigera. Bioassays were used to determine whether or not the H. armigera samples acquired from the wild were resistant to Bt. In order to perform the experiment, a total of 250 Bt cotton field-collected larvae and 25 Chickpea field-collected larvae were sampled from the laboratory culture.

### F1 Generation

Fresh Bt and non-Bt cotton leaves were subjected to a bioassay with H. armigera in the lab. Bioassays were performed on neonates acquired by rearing larvae on Bt cotton leaves and bolls until they emerged as adults. Protocol that was followed to carry out the bioassay. From February 3rd to February 9th, 2020, bioassays were performed using freshly hatched (one day old) H. armigera larvae on 250 Bt hybrid leaves and 25 non-Bt hybrid leaves, respectively.

**F2 Generation**

The screened larvae obtained from the F1 generation were reared in the laboratory until adult emergence and then adults were paired & kept in oviposition cage. The neonates obtained from F1 female were considered as F2 generation and used for bioassay. Leaves of Bt and non-Bt were brought to the laboratory and the newly-hatched (one day old) H. armigera larvae 25 larvae each on Bt and Non-Bt, were used in bioassay that was conducted from 15th to 21st March 2020. Observations on the weight of surviving larvae and mortality were recorded on 7th day after release.

**3.3 Population dynamics of h. armigera**

**3.3.1 Bt cotton**

The prevalence of H. armigera was investigated in the Bt cotton fields in the Vadodara district of Gujarat. Beginning in the blooming phase, weekly checks for H. armigera larvae in Bt cotton fields will continue until January 2020. Surveillance was focused on the worst-managed Bt cotton crops or stressed-out fields. Using a plant inspection approach, the larval population was monitored weekly across 100 plants in five different fields.

**3.3.2 Chickpea**

A field research was conducted in farmers' fields in Gudel and Tamsa in Anand, Gujarat in 2018–19 and 2019–20 to assess the impact of H. armigera on chickpea, Cicer arietinum L., across the several seasons. The best agronomic practises for this area were used to grow the crops. Larvae of H. armigera were monitored for their emergence and population growth every week. The number of Helicoverpa larvae per plant per week was recorded from the first week of November 2018 through the first week of March 2019, and from the first week of November 2019 through the first week of March 2020, using a random sample of twenty plants from the field.

**3.3.3 Natural regulation of H. armigera**

The study was performed in Anand between 2018 and 2020 to count H. armigera larvae and identify possible parasitoids on chickpea. There was a comprehensive census of larvae and parasitoids on the chickpea plants. Between fifty and one hundred larvae were taken from the wild and maintained in the lab on an artificial diet until they emerged as adults (from the unparasitized larvae) and parasitoids (from the parasitized larvae). To keep track of parasitized larvae and the appearance of parasitoids, observation must be performed every other day.

# 4. Results

## 4.1 Screening and isolation of bt resistance in h. armigera (Hübner)

The H. armigera larva was collected via numerous trips to the Bt cotton fields in the Karjan area of the Vadodara district of Gujarat. During the 2019-2020 survey season, larvae resistant to Bt cotton were collected from 105 cotton fields once a week. From the first week of September 2019 through the final week of January 2020, H. armigera was collected from Bt cotton fields. Larvae of H. armigera were collected from poorly maintained Bt cotton fields that were in the flowering/fruiting stage. The resistant H. armigera larvae were recovered from four distinct Bt cotton fields out of a total of 105 crops examined. A total of 71 Bt-resistant H. armigera larvae were collected during the survey, and the study also documented the types of plant tissues (younger leaves, squares, flowers, and bolls) on which the larvae were found eating. H. armigera larvae were most often seen on bolls (60.56 percent), followed by tender leaves (16.90 percent), squares (14.0 percent), flowers (5.6 percent), and older leaves (2.8 percent).

The survey was done weekly throughout 2019 and 2020 to collect H. armigera larvae from vulnerable populations in okra, chickpea, and other host crops. From the third week of September 2019 through the first week of March 2020, the survey was open for responses. During the survey, several plant components and 500-750 H. armigera larvae were brought back to the lab.

**Table 4.1: Hemlock budworm (H. armigera) samples were gathered from Bt cotton crops in Karjan Taluka, Vadodara district.**

| Sr.No. | Dateofcollection | No.of*H.armigera*larvae | Place |
|---|---|---|---|
| 1 | 09.09.2019 | 02 | Vemardi |
| 2 | 16.09.2019 | 02 | Karjan |
| 3 | 22.10.2019 | 05 | Kambola |
| 4 | 21.11.2019 | 04 | Miyagam |

| | | | |
|---|---|---|---|
| 5 | 16.12.2019 | 06 | Mangrol |
| 6 | 20.12.2019 | 15 | Mangrol |
| 7 | 26.12.2019 | 32 | Mangrol |
| 8 | 28.01.2020 | 05 | Mangrol |

**F1 Generation**

In bioassays using Bt and non-Bt cotton leaves, the average weight of surviving H. armigera larvae was 10.51 1.40 mg on Bt cotton and 23.07 3.38 mg on non-Bt cotton (Table 4.2). Mortality for the F1 generation was 29.60% and 4.00%, respectively.

**F2 Generation**

Adults were coupled off and housed in an oviposition cage after being raised from screened larva collected from the F1 generation. For the purpose of biotesting, F2 generation neonates were employed. The average weight of surviving larvae was 10.87 1.30 mg on Bt cotton and 24.52 1.67mg on non-Bt cotton for F2 generation (Table 4.2), and mortality of H. armigera on Bt and non-Bt cotton leaves was 28.00% and 8.00%, respectively.

**Table 4.2: Larval bioassay of Bt and non-Bt cotton leaves against H. armigera**

| **Treatment** | **Totalno.larvaeused (n)** | **Mortality (%)** | **Wt. of survivingindividuals(mg) Mean±SD** |
|---|---|---|---|
| **$F_1$Generation** | | | |
| *Bt*Cotton | 250 | 29.60 | 10.51± 1.40 |
| Non-*Bt* Cotton | 25 | 4.00 | 23.07± 3.38 |
| **$F_2$Generation** | | | |
| *Bt*Cotton | 25 | 28.00 | 10.87± 1.30 |
| Non-*Bt* Cotton | 25 | 8.00 | 24.52± 1.67 |

Bioassay findings indicated that the first generation of H. armigera on Bt cotton had a greater death rate than the second generation. The H. armigera larval survival rate was substantially greater in the non-Bt cotton treatment compared to the Bt cotton treatment (P 0.01).

**4.2 Evaluation of fitness cost of bt resistance in h. Armigera on NON-Bt cotton**

According to new research comparing the fitness costs of Bt-resistant and -susceptible H. armigera populations on non-Bt cotton, the Bt-resistant population of H. armigera on non-Bt cotton recorded the longest egg, larval, and pupal periods, while the H. armigera population on non-Bt cotton had the shortest adult life span.

Tables 4.3 provide the calculated findings of fitness costs of different biological characteristics of Bt resistant populations of H. armigera on non-Bt cotton. The results showed that the length and width of newly laid eggs ranged from 0.44 to 0.52 mm and 0.48 to 0.54 mm, on average 0.47 0.03 mm and 0.51 0.02 mm, and that Bt-resistant and -susceptible populations of H. armigera had lengths and widths of 0.50 0.04 mm and 0.52 0.04 mm, respectively. Table 4.4 displays the fitness cost of varying egg length and width.

The findings show that the average incubation time for the Bt-resistant H. armigera population was 3.55 0.51 days, whereas the average incubation time for the susceptible H. armigera population was 3.45 0.51 days. Incubation time has a 2.90 percentage point negative impact on fitness. According to the results of the current studies, the hatchability of the Bt-resistant population of H. armigera varied from 30.28 to 36.84 percent, with an average of 33.59 3.03, whereas that of the susceptible population ranged from 35.98 to 42.44 percent, with an average of 38.91 3.07. Table 4.3 shows that the fitness cost of hatching was 13.67 percent.

The head capsule was big for its kind and ranged in colour from almost black to a dark brown. Table 4.6 shows that the head capsule length and width of the Bt-resistant and susceptible populations of H. armigera ranged from 0.16 to 0.33 mm (0.24 0.06 mm) and 0.19 to 0.35 mm (0.28 0.05 mm), 0.17 to 0.31 mm (0.25 0.05), and 0.21 to 0.39 (0.29 0.06 mm), respectively.Table 4.4 summarises the data and shows that the first instar larvae of the Bt-resistant and susceptible populations of H. armigera lived for a mean of 2.55 0.51 days and 1.85 0.37 days, respectively.Table 4.6 shows that the weight of H. armigera larvae in their first instar varied from 2.80 to 3.50 mg, with an average of 3.11 0.23 mg, for the Bt-resistant population, and from 4.20 to 6.00 mg, with an average of 5.32 0.54 mg, for the susceptible population. Table 4.4 shows that the fitness cost of a longer first instar larval stage was 37.84%, while a heavier first instar larval stage cost 41.54%, and the fitness cost of a longer and wider first instar larval head capsule was 4.00 and 3.45%, respectively.

In the study of Bt-resistant and -susceptible populations of H. armigera, the length of the head capsule varied from 0.36 to 0.44 mm (0.41 0.03 mm) and the width from 0.39 to 0.46 mm (0.42 0.02 mm) and 0.42 to 0.55 mm (0.48 0.04 mm), respectively (Table 4.6).Table 4.4 shows that the second instar larval stage of both the Bt-resistant and susceptible H. armigera populations took between 2 and 3 days (2.60 0.50 days) to complete.Second instar H. armigera larval weights varied from 8.50 to 12.20 mg (average: 10.14 1.88 mg) in the Bt resistant population and from 26.60 to 31.50 mg (average: 28.81 1.65 mg) in the susceptible population (Table 4.6).Weight, head capsule length, and breadth all increased by 64.80%, 12.77%, and 12.50% in the second instar larva compared to the first (Table 4.5).

Compact and see-through with faint brown markings, the head capsule was less voluminous. Third instar H. armigera larvae's head capsules ranged in length from 0.54 to 0.61 mm (0.56 0.02 mm), width from 0.57 to 0.65 mm (0.59 0.04 mm), and breadth from 0.61 to 0.72 mm (0.66 0.03 mm) for the Bt-resistant and susceptible populations, respectively (Table 4.6).Third instar development in the Bt-resistant H. armigera population took around 3.50 0.51 days, whereas in the susceptible population it took about 2.50 0.51 days (Table 4.4).Third instar H. armigera larval weights varied from 42.00 to 49.30 mg (average = 45.76 2.37 mg) in the Bt resistant population and from 65.80 to 76.10 mg (average = 70.94 3.40 mg) in the susceptible population (Table 4.6). Tables 4.3 and 4.5 show that the costs to fitness associated with third-stage larval longevity, weight, and head capsule length and breadth are 40.00, 35.49, 11.11, and 10.61%, respectively.

The head capsule length and width of H. armigera fourth instar larvae ranged from 1.15 to 1.38 mm and 1.16 to 1.44 mm, respectively, with averages of 1.23 0.09 and 1.25 0.11 mm (Table 4.6). The Bt resistant population averaged 1.29 0.08 mm and the susceptible population averaged 1.35 0.09 mm. Fourth instar H. armigera larvae lived for 3.55 0.51 days when exposed to Bt and 3.40 0.50 days when exposed to a sensitive population (Table 4.4).Larval weights for Bt-resistant and susceptible H. armigera populations varied from 98.10 to 115.20 mg (105.61 5.30 mg) and 106.40 to 127.00 mg (115.75 6.25 mg), respectively (Table 4.6).Having a longer and heavier larva during the fourth instar reduced

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fitness by 4.41 and 8.76 percent, respectively. The costs to fitness associated with a longer and wider fourth-instar head capsule are 4.65 and 7.41%, respectively (Tables 4.3 and 4.5).

Head capsule length and width varied between 1.65 and 1.78 mm (average: 1.70 0.06 mm) in the Bt-resistant population of H. armigera and 1.71 and 1.80 mm (1.78 0.02 and 1.80 0.05 mm), 1.73 and 1.87 mm (1.80 0.05 and 1.81 and 2.06 mm (1.91 0.08 mm), respectively (Table 4.6).Fifth instar H. armigera larvae lived between 4 and 5 days on average (4.40 0.50 days) in the Bt-resistant population and between 3 and 5 days on average (4.25 0.72) in the susceptible population (Table 4.4). Both the Bt-resistant and -susceptible H. armigera populations had fifth-instar larvae weighing between 213.00 and 236.50 mg (average: 227.90 5.87 mg) and between 242.20 and 258.00 mg (average: 249.44 4.30 mg), respectively (Table 4.6). The costs to fitness associated with living longer and weighing more as a fifth-stage larva were 3.53% and 8.64%, respectively. Both the length and breadth of the fifth instar's head capsule had negative effects on fitness, with 5.56 and 6.81%, respectively (Tables 4.3 and 4.5).

The length of the head capsule ranged from 1.82 to 1.89 millimetres (1.86 0.02 millimetres), the width from 1.92 to 2.38 millimetres (1.89 0.68 millimetres), and the height from 2.08 to 2.65 millimetres (2.35 0.19 millimetres) in the Bt-resistant and susceptible populations of H. armigera, respectively (Table 4.6).Sixth instar larval development time was recorded to be between 4 and 5 days (4.50 0.51 days) for both Bt-resistant and susceptible H. armigera populations (Table 4.4).Sixth instar H. armigera larval weights varied from 323.40 to 386.50 mg (average: 355.19 18.11 mg) in the Bt-resistant population and from 362.60 to 400.00 mg (average: 379.54 17.28 mg) in the susceptible population (Table 4.6). Bt-resistant H. armigera larvae were heavier and lived longer, albeit at a loss of 2.27 percent of their fitness.The sixth instar's head capsule length and breadth had fitness costs of 2.62 and 19.57 percent, respectively (Tables 4.3 and 4.5).

The data analysis showed that the overall larval development time of H. armigera ranged from 21 to 28 days (24.50 4.95 days) for the Bt resistant population and from 18 to 26 days (22.0 5.66) for the susceptible population. The current findings showed that Bt-resistant H. armigera spent more time as larvae on non-Bt cotton than the susceptible population. The existence of a fitness penalty, estimated at 11.36 percent in Table 3.4, may explain why resistant populations on non-Bt cotton have longer larval stages than susceptible populations.

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**Table 4.3: The effect of Bt resistance on H. armigera (n=20) fitness as measured by several biological parameters**

| Sr. No. | Developmental stages(Days) | Mean± S.D. | | Fitnesscost(%) $[(F_R-F_S/F_S)]\times100$ |
|---|---|---|---|---|
| | | $F_R$ | $F_S$ | |
| 1 | Eggperiod | 3.55± 0.51 | 3.45± 0.51 | 2.90 |
| 2 | Ilarvalinstar | 2.55± 0.51 | 1.85± 0.37 | 37.84 |
| 3 | IIlarvalinstar | 2.60± 0.50 | 2.50± 0.51 | 4.00 |
| 4 | IIIlarvalinstar | 3.50± 0.51 | 2.50± 0.51 | 40.00 |
| 5 | IV larvalinstar | 3.55±0.51 | 3.40±0.50 | 4.41 |
| 6 | V larvalinstar | 4.40± 0.50 | 4.25±0.72 | 3.53 |
| 7 | VIlarvalinstar | 4.50± 0.51 | 4.40±0.50 | 2.27 |
| 8 | Totallarvalperiod | 24.5± 4.95 | 22.0± 5.66 | 11.36 |
| 9 | Pre-pupalperiod | 1.55± 0.51 | 1.50± 0.51 | 3.33 |
| 10 | Malepupal period | 9.71± 1.11 | 9.38± 0.52 | 3.52 |
| 11 | Female pupalperiod | 9.15± 0.80 | 8.54± 0.52 | 7.14 |
| 12 | Malelongevity | 7.50± 0.93 | 7.86± 1.07 | 4.80 |
| 13 | Female longevity | 8.83± 0.72 | 8.46± 1.05 | 4.37 |
| 14 | Totalperiod(Male) | 42.5± 7.78 | 40.5±10.61 | 4.94 |
| 15 | Totalperiod(Female) | 43.5±7.78 | 41.0±9.90 | 6.10 |
| 16 | Pre-ovipositionperiod | 2.42± 0.51 | 2.23± 0.73 | 8.52 |
| 17 | Oviposition period | 4.71± 0.76 | 5.85±0.80 | -19.49 |
| 18 | Post-ovipositionperiod | 1.54± 0.52 | 2.08± 0.79 | -25.96 |
| 19 | Fecundity (Eggs/female) | 42.94± 22.51 | 44.02± 12.21 | 2.45 |
| 20 | Sexratio(Male:Female) (n=5) | 1:1.6 | 1:1.83 | 12.57 |
| 21 | Hatching% | 33.59± 3.03 | 38.91± 3.07 | 13.67 |
| 22 | Growthindex | 3.36 | 4.70 | 28.51 |
| 23 | Survivalindex | 0.65 | 0.85 | 23.53 |

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**Table 4.4: The effect of Bt resistance on fitness was evaluated using twenty samples of H. armigera.**

| Sr. No. | Particulars | Mean± S.D | | Fitness cost (%)[($F_S$-$F_R$/$F_S$)]×100 |
|---|---|---|---|---|
| | | $F_R$ | $F_S$ | |
| | | **Measurementslength (mm)** | | |
| 1 | Egg | 0.47± 0.03 | 0.50± 0.04 | 6.00 |
| 2 | IInstar | 1.39± 0.05 | 1.42± 0.07 | 2.11 |
| 3 | IIInstar | 2.98±0.12 | 3.25±0.36 | 8.31 |
| 4 | IIIInstar | 10.14± 0.74 | 10.73± 1.73 | 5.50 |
| 5 | IV Instar | 20.05± 1.18 | 21.90± 1.55 | 8.45 |
| 6 | V Instar | 31.08± 1.34 | 32.34± 2.15 | 3.90 |
| 7 | VIInstar | 37.42± 1.26 | 42.71± 1.31 | 12.39 |
| 8 | Pre-pupa | 22.30± 1.33 | 23.89± 1.53 | 6.66 |
| 9 | Pupa | 19.33± 0.43 | 21.17± 1.13 | 8.69 |
| 10 | Malepupa | 14.97± 2.21 | 15.69± 0.39 | 4.59 |
| 11 | Female pupa | 18.47± 0.41 | 19.68± 0.97 | 6.15 |
| 12 | Maleadult | 15.82± 0.37 | 16.67± 0.71 | 5.10 |
| 13 | Female adult | 18.91± 1.99 | 19.62± 2.01 | 3.62 |
| | | **Measurementswidth (mm)** | | |
| 1 | Egg | 0.51± 0.02 | 0.52± 0.04 | 1.92 |
| 2 | IInstar | 0.52± 0.02 | 0.54± 0.02 | 3.70 |
| 3 | IIInstar | 0.70± 0.04 | 0.76± 0.03 | 7.89 |
| 4 | IIIInstar | 1.95± 0.06 | 2.32± 0.09 | 15.95 |
| 5 | IV Instar | 2.57± 0.12 | 3.20± 0.05 | 19.69 |
| 6 | V Instar | 3.97± 0.43 | 4.79± 0.27 | 17.12 |
| 7 | VIInstar | 5.42± 0.26 | 6.54± 0.61 | 17.13 |
| 8 | Pre-pupa | 4.40± 0.08 | 4.87± 0.03 | 9.65 |
| 9 | Pupa | 5.22± 0.27 | 5.94± 0.06 | 12.12 |
| 10 | Malepupa | 2.67± 0.03 | 3.60± 0.07 | 25.83 |
| 11 | Female pupa | 3.81± 0.07 | 4.90± 0.04 | 22.24 |
| 12 | Maleadult | 29.14± 1.19 | 31.38± 1.00 | 7.14 |
| 13 | Female adult | 29.08± 1.17 | 32.01± 2.13 | 9.15 |

**Table 4.5: Weight gain resistance in Bt-fit H. armigera (n=20): the cost to fitness.**

| Sr. | | Mean± S.D | Fitness cost |
|---|---|---|---|

| No. | Particulars | $F_R$ | $F_S$ | (%)[($F_S$-$F_R$/$F_S$)]×100 |
|---|---|---|---|---|
| **Meanweight(mg)** | | | | |
| 1 | IInstar | 3.11± 0.23 | 5.32± 0.54 | 41.54 |
| 2 | IIInstar | 10.14± 1.88 | 28.81± 1.65 | 64.80 |
| 3 | IIIInstar | 45.76± 2.37 | 70.94± 3.40 | 35.49 |
| 4 | IV Instar | 105.61± 5.30 | 115.75± 6.25 | 8.76 |
| 5 | V Instar | 227.90± 5.87 | 249.44± 4.30 | 8.64 |
| 6 | VIInstar | 355.19± 18.11 | 379.54± 17.28 | 6.42 |
| 7 | Pupa | 337.30± 13.25 | 374.55± 6.52 | 8.83 |
| 8 | Malepupa | 321.88± 12.27 | 353.06± 4.10 | 10.86 |
| 9 | Female pupa | 310.54± 1.72 | 348.38± 2.23 | 6.53 |
| **Meanheadcapsulelength(mm)** | | | | |
| 1 | IInstar | 0.24± 0.06 | 0.25± 0.05 | 4.00 |
| 2 | IIInstar | 0.41± 0.03 | 0.47± 0.04 | 12.77 |
| 3 | IIIInstar | 0.56± 0.02 | 0.63± 0.03 | 11.11 |
| 4 | IV Instar | 1.23± 0.09 | 1.29± 0.08 | 4.65 |
| 5 | V Instar | 1.70± 0.06 | 1.80± 0.05 | 5.56 |
| 6 | VIInstar | 1.86± 0.02 | 1.91± 0.04 | 2.62 |
| **Meanhead capsulewidth(mm)** | | | | |
| 1 | IInstar | 0.28± 0.05 | 0.29± 0.06 | 3.45 |
| 2 | IIInstar | 0.42± 0.02 | 0.48± 0.04 | 12.50 |
| 3 | IIIInstar | 0.59± 0.04 | 0.66± 0.03 | 10.61 |
| 4 | IV Instar | 1.25± 0.11 | 1.35± 0.09 | 7.41 |
| 5 | V Instar | 1.78± 0.02 | 1.91± 0.08 | 6.81 |
| 6 | VIInstar | 1.89± 0.68 | 2.35± 0.19 | 19.57 |

A population's death toll may be easily summarised in a life table. For each age group, life details the number of fatalities, the number of survivors, the mortality rate, and the likelihood of continued existence. A life table is a useful tool for analysing the fluctuations in insect populations throughout their many phases of development. Out of 50 eggs, only 13 from the Bt-resistant population and 17 from the susceptible group made it to adulthood (Table 4.7).

Mortality rates in the egg stage were 60.00%, in the larval stage 13.33%, and in the pupal stage 5.56% for both the Bt resistant and susceptible populations of H. armigera. Mortality was highest across Bt-resistant and -susceptible H. armigera populations during the first instar of larval development (10% and 5%, respectively), and lowest among the sixth instar (5.88%). The Bt-resistant population of H. armigera had a survival fraction (Sx) of 1.00 on stages III, IV, and V and 0.90 on stage I, whereas the susceptible population had a Sx of 1.00 on stages II, III, IV, V, and VI and 0.95 on stage I. Mortality to survival ratios showed that the highest value was observed during the egg stage (1.50) and the lowest occurred during the third, fourth, and fifth instars (0.00) for both the Bt-resistant and susceptible populations of H. armigera.

Births at age x (mx) and the probability of female survival (lx) were calculated using the age-specific fecundity formula. The findings showed that the pre-oviposition phase for the Bt-resistant and susceptible populations of H. armigera was between the 31st and 33rd day and the 28th and 30th of the crucial age. Females of the Bt-resistant H. armigera population laid their first eggs on day 34 (mx=14.50) and continued to do so until day 38 (mx=62.50), while those of the Bt-susceptible H. armigera population did so on day 31 (mx=26.45) and laid their first eggs on day 36 (mx=47.50), respectively. Female mortality began on day 4, post-adult emergence, or day 35 of crucial age (lx=0.88), and gradually rose beyond that point, as shown by a decline in lx values.

A population of Bt-resistant H. armigera doubles in 5.50 days, while a population of Bt-susceptible H. armigera doubles in 4.61 days. The intrinsic rate of natural increase in number (rm) was 0.126 and 0.151 females/female/day for the Bt-resistant and susceptible populations, respectively. Under these circumstances, the Bt-resistant population of H. armigera may grow by 1.09 times per week, whereas the susceptible population can multiply by 1.80 times per week. Potential fecundity was calculated to be 214.69 and 267.58 eggs/female for the Bt-resistant and susceptible populations of H. armigera, respectively, in the F2 generation. Net reproductive rate (Ro), mean generation time (Tc), intrinsic rate of natural increase in number (rm), finite rate of increase in number (), time to double the population, time to multiply the population by two each week, hypothetical F2 females, and potential fecundity were all negative for the population (Table 4.6).

**Table 4.6: H. armigera Bt-resistance population age distribution on non-Bt cotton (n=50).**

| Pivotal ageindays(x) | Lx | x+1 | $-r_m$*(x+1) | $e^{-rm (x+1)}$ | $Lx.e^{-rm(x+1)}$ | Contribution(%) | |
|---|---|---|---|---|---|---|---|
| 0 | 1.00 | 1 | -0.0126 | 0.9874 | 0.9874 | 3.4087 | 10.0982 (Egg) |
| 1 | 1.00 | 2 | -0.0253 | 0.9750 | 0.9750 | 3.3659 | |
| 2 | 1.00 | 3 | -0.0379 | 0.9628 | 0.9628 | 3.3236 | |
| 3 | 1.00 | 4 | -0.0506 | 0.9507 | 0.9507 | 3.2818 | |
| 4 | 1.00 | 5 | -0.0632 | 0.9387 | 0.9387 | 3.2406 | 45.1255 (Larva) |
| 5 | 1.00 | 6 | -0.0759 | 0.9270 | 0.9270 | 3.1999 | |
| 6 | 1.00 | 7 | -0.0885 | 0.9153 | 0.9153 | 3.1597 | |
| 7 | 1.00 | 8 | -0.1011 | 0.9038 | 0.9038 | 3.1200 | |
| 8 | 1.00 | 9 | -0.1138 | 0.8925 | 0.8925 | 3.0808 | |
| 9 | 1.00 | 10 | -0.1264 | 0.8812 | 0.8812 | 3.0421 | |
| 10 | 1.00 | 11 | -0.1391 | 0.8702 | 0.8702 | 3.0039 | |
| 11 | 1.00 | 12 | -0.1517 | 0.8592 | 0.8592 | 2.9661 | |
| 12 | 1.00 | 13 | -0.1644 | 0.8484 | 0.8484 | 2.9289 | |
| 13 | 1.00 | 14 | -0.1770 | 0.8378 | 0.8378 | 2.8921 | |
| 14 | 1.00 | 15 | -0.1896 | 0.8273 | 0.8273 | 2.8558 | |
| 15 | 1.00 | 16 | -0.2023 | 0.8169 | 0.8169 | 2.8199 | |
| 16 | 1.00 | 17 | -0.2149 | 0.8066 | 0.8066 | 2.7845 | |
| 17 | 1.00 | 18 | -0.2276 | 0.7965 | 0.7965 | 2.7495 | |
| 18 | 1.00 | 19 | -0.2402 | 0.7865 | 0.7865 | 2.7149 | |
| 19 | 1.00 | 20 | -0.2528 | 0.7766 | 0.7766 | 2.6808 | |
| 20 | 1.00 | 21 | -0.2655 | 0.7668 | 0.7668 | 2.6472 | |
| 21 | 1.00 | 22 | -0.2781 | 0.7572 | 0.7572 | 2.6139 | |
| 22 | 1.00 | 23 | -0.2908 | 0.7477 | 0.7477 | 2.5811 | |
| 23 | 1.00 | 24 | -0.3034 | 0.7383 | 0.7383 | 2.5486 | |
| 24 | 1.00 | 25 | -0.3161 | 0.7290 | 0.7290 | 2.5166 | |
| 25 | 1.00 | 26 | -0.3287 | 0.7199 | 0.7199 | 2.4850 | 20.7881 (Pupa) |
| 26 | 1.00 | 27 | -0.3413 | 0.7108 | 0.7108 | 2.4538 | |
| 27 | 1.00 | 28 | -0.3540 | 0.7019 | 0.7019 | 2.4230 | |
| 28 | 1.00 | 29 | -0.3666 | 0.6931 | 0.6931 | 2.3925 | |
| 29 | 1.00 | 30 | -0.3793 | 0.6844 | 0.6844 | 2.3625 | |
| 30 | 1.00 | 31 | -0.3919 | 0.6758 | 0.6758 | 2.3328 | |
| 31 | 1.00 | 32 | -0.4046 | 0.6673 | 0.6673 | 2.3035 | |
| 32 | 1.00 | 33 | -0.4172 | 0.6589 | 0.6589 | 2.2745 | |
| 33 | 1.00 | 34 | -0.4298 | 0.6506 | 0.6506 | 2.2460 | |
| 34 | 0.94 | 35 | -0.4425 | 0.6424 | 0.6023 | 2.0791 | 23.9882 (Adult) |
| 35 | 0.75 | 36 | -0.4551 | 0.6344 | 0.4758 | 1.6424 | |
| 36 | 0.44 | 37 | -0.4678 | 0.6264 | 0.2740 | 0.9460 | |
| 37 | 0.19 | 38 | -0.4804 | 0.6185 | 0.1160 | 0.4004 | |
| 38 | 0.06 | 39 | -0.4931 | 0.6108 | 0.0382 | 0.1318 | |
| 39 | 0.00 | 40 | -0.5057 | 0.6031 | 0.0000 | 0.0000 | |
| | | | | | | 100 | 100 |

**Table 4.7: The population parameter cost of Bt resistance in H. armigera's fitness**

| Sr. No. | Parameters | Formula | $F_R$ | $F_S$ | Fitnesscost(%) $[(F_S-F_R/F_S)]\times100$ |
|---|---|---|---|---|---|
| 1 | Netreproductiverate(No./female/lifetime) | $R_0=\Sigma lxmx$ | 90.56 | 138.64 | 34.68 |
| 2 | Weekly multiplicationof population(times) | $(\lambda)^7$ | 1.09 | 1.80 | 39.44 |
| 3 | Hypothetical$F_2$females | $(Ro)^2$ | 8201.57 | 19220.04 | 57.33 |
| 4 | Potential fecundity(PF) | $\Sigma mx$ | 214.69 | 267.58 | 19.77 |
| | | | $F_R$ | $F_S$ | Fitnesscost(%) $[(F_R-F_S/F_S)]\times100$ |
| 1 | Mean length generation time(days) | $T_C=\Sigma x.lx.mx/Ro$ | 35.73 | 32.77 | 9.03 |
| 2 | Innatecapacity forincreasein numbers(Females/female/day) | $rm=log_eRo/Tc$ | 0.126 | 0.151 | -16.56 |
| 3 | rm=intrinsicrateofnaturalincrease(Females/female/day) | $\Sigma e7^{-rmx}.Lxmx$ | 0.126 | 0.151 | -16.56 |
| 4 | Finiterateofincreasein number(Females/female/day) | Antilog $e^{rm}$ | 1.01 | 1.09 | -7.34 |
| 5 | Populationdoublingtime(DT)(Days) | =loge2/rm | 5.50 | 4.61 | 19.31 |

**Overall fitness cost (C) and the resistance ratio (Rr)**

The present study found that on non-Bt cotton, the intrinsic rate of population increase (rm), the cost of fecundity (CFec), the cost of copulation (CRc), the cost of pupal weight (CWp), and the cost of survival from the sixth instar larvae to pupae (6.38) were all highest in the susceptible population and lowest in the resistant population..

**Table 4.8: Overall expense for fitness (C)**

| Sr. No. | Particulars | Meanvalues | | Overall fitness cost (C)(%) $[(r_mS-r_mR)/r_mS]\times100$ |
|---|---|---|---|---|
| | | $r_mR$ | $r_mS$ | |
| 1 | Intrinsicrateof populationincrease | 0.15 | 0.13 | 16.22 |

## 5. Conclusion

Transgenic crops that produce Bacillus thuringiensis toxins (Bt) have been widely planted for over two decades. These toxins are effective against lepidopteran pests. Large-scale

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production of Bt crops presents a number of challenges, including the management of resistance. Fitness costs associated with Bt resistance, i.e. lower fitness in resistant insects than in susceptible ones in the absence of Bt toxins, can further delay the development of resistance by selecting against Bt-resista. The present study found that Bt resistant H. armigera larvae grows and develops more slowly than the Bt susceptible when reared on absence of Bt toxin (non-Bt cotton).

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