Natural Enemies in different *Bt* Cotton hybrids under Protected and Unprotected conditions

ABSTRACT

A field experiment was conducted to analyze the population dynamics of natural enemies including *Chrysoperla* spp., coccinellids, and spiders, in Bt cotton under both protected and unprotected conditions. The results indicated that the population of natural enemies was significantly higher under unprotected conditions compared to protected fields, where insecticides were applied. Among the tested hybrids, G. Cot. Hy.10 BG recorded the highest predator activity, showing greater population of Chrysoperla, coccinellids, and spiders in unprotected conditions. In contrast, the lowest predator activity was observed in RCH 2 BG , while Ajeet 155 BG and ATM BG had minimal natural enemy presence. These findings highlight the importance of unprotected conditions in preserving natural enemies and suggest that certain Bt hybrids, such as G. Cot. Hy.10 BG , may better support predator populations. The study emphasizes the need to integrate natural enemies into pest management strategies for sustainable cotton production. Predators of pests, coccinellids and *Chrysoperla were* observed from 33rd to 52nd whereas spider population was observed from the 31st to 52nd SW under unprotected conditions.

Keywords: *Bt* cotton, Natural enemies, Protected condition, Unprotected condition

Introduction

Cotton is commonly known as "White Gold," and it is a crucial cash crop in India, significantly contributing to the nation's economy. It supports the livelihoods of over 60 million people through its role in agriculture, processing, and its extensive use in the textile industry [1]. Cotton cultivation in India faces significant challenges from 162 species of insect pests, which infest the crop throughout its growth cycle, from planting to maturity [2]. Cotton hosts both tissue borers and sap-feeding insects, which can lead to a 30-90% reduction in yield. Key sucking pests include the aphid *Aphis gossypii* (Glover), the leafhopper *Amrasca biguttula biguttula* (Ishida), the thrips *Thrips tabaci* (Lind.), and the whitefly *Bemisia tabaci* (Genn.), which are of particular concern. By the early 1990s, sucking pests, especially leafhoppers and whiteflies, began causing severe damage when not adequately controlled. The use of imidacloprid-treated seeds became a common strategy to manage early-season infestations; however, this approach has further intensified sucking pest issues in several crops [3]. Cotton growers are increasingly using a variety of systemic pesticides to combat sucking insect pests, but these efforts have largely been unsuccessful. This is likely due to the development of resistance in these pests to many commonly used synthetic insecticides [4]. In addition, the excessive use of broad-spectrum insecticides raises concerns about environmental pollution and their unintended negative effects on beneficial organisms, such as insect predators (ants, coccinellid beetles, and green lacewings) and parasitoids like wasps [5]. Therefore, the present investigation was performed with the natural enemies population under protected and unprotected conditions to know the impact of chemical insecticides on natural enemies population.

Materials and Methods

The present investigation on the Natural enemies in *Bt* cotton was conducted under field conditions at Main Cotton Research Station, Navsari Agricultural University, Surat

(Gujarat) during *Kharif*, 2023-24. Four Bt cotton hybrids (ATM BG II, Ajeet 155 BG II, RCH 2 BG II, G. Cot. Hy. 10 BG II) were evaluated under protected and unprotected condition. The study involved monitoring the population of key natural enemies, including *Chrysoperla* (larvae), coccinellids (both larvae and adults) and spiders (adults), on Bt cotton plants under two distinct management regimes: protected and unprotected conditions. To obtain representative data, five plants were randomly selected from each treatment plot for close observation. The protected plots received regular insecticide applications (Flonicamid 50 WG 0.015%, Thiamethoxam 25 WG 0.0075%, Buprofezin 25 SC 0.05%, Diafenthiuron 50 WP 0.05%) as part of a conventional pest management strategy, while the unprotected plots were left without any chemical treatments, allowing for natural pest and predator interactions. The selected plants were thoroughly inspected at regular intervals, and the number of natural enemies present was recorded. Among them, *Chrysoperla* larvae known for their role as effective biological control agents due to their voracious predation on aphids and other soft-bodied pests, were carefully counted on the plants. Coccinellids which included both larvae and adults of ladybird beetles were also noted and their importance in controlling a variety of insect pests such as aphids, scale insects, and mites. Adult spiders as generalist predators, were recorded as well due to their role in capturing a wide range of insect prey including flies, moths, and beetles. By conducting observations across both protected and unprotected conditions, the study aimed to compare how the different pest control strategies affected the presence and activity of these natural enemies. The random selection of plants ensured that the data collected represented the overall population trends in the field, allowing for a robust analysis of natural enemy dynamics in relation to pest control practices in Bt cotton. This methodology provided insight into the ecological balance between pests and their natural enemies, contributing valuable information for future pest management strategies.

Result and discussion

Chrysoperla:

The pooled data on the larval population of Chrysoperla, was presented in Table.1 demonstrates a significant variation between the protected and unprotected plots. Specifically, a higher population of Chrysoperla larvae was consistently recorded in the unprotected plots with an average of 0.62 larvae per plant compared to protected plots, which had a lower average of 0.42 larvae per plant. This significant difference suggests that the absence of insecticide applications in the unprotected plots favoured the survival and activity of Chrysoperla larvae, allowing for a more robust population. Interestingly, while the protection levels had a notable impact on the *Chrysoperla* larval population, the different Bt cotton hybrids under study did not show a statistically significant effect on larval numbers. Regardless of the hybrid being cultivated, the population levels of Chrysoperla larvae remained consistent, indicating that the variation in natural enemy population was largely driven by the pest management practices rather than the specific characteristics of the Bt hybrids themselves. Moreover, the interaction between the hybrids and the protection levels did not show any significant differences. This implies that the influence of protection strategies on *Chrysoperla* population was independent of the cotton hybrids planted, further emphasizing that the use of chemical inputs played a dominant role in affecting the population dynamics of this important predator.

Table 1: Impact of hybrids and protection on the population of *Chrysoperla* in cotton (Pooled)

Treatments	No. of *Chrysoperla* larvae/plant				
Main\ Sub plot	**H_1 (ATM BGII)**	**H_2 (Ajeet 155 BGII)**	**H_3 (RCH 2 BGII)**	**H_4 (G.Cot.Hy. 10 BGII)**	**Mean**
P_1 (Protected)	0.96 (0.42)	0.97 (0.45)	0.94 (0.38)	0.97 (0.45)	0.96[b] (0.42)
P_2 (Unprotected)	1.05 (0.60)	1.07 (0.64)	1.05 (0.60)	1.07 (0.64)	1.06[a] (0.62)
Mean	1.01 (0.52)	1.02 (0.54)	1.00 (0.50)	1.02 (0.54)	1.01 (0.52)
Interactions	**Protection (P)**	**Hybrid (H)**	**P x H**	**C.V. (%) Main**	
S. Em. ±	0.02	0.008	0.01	7.94	
C. D. at 5%	0.06	NS	NS		
	Period (Y)	**P x Y**	**H x Y**	**P x H x Y**	**C.V. (%) Sub**
S. Em. ±	0.01	0.02	0.02	0.03	7.45
C. D. at 5%	0.04	0.05	NS	NS	

Notes:
Figures in parentheses are retransformed values, those outside are $\sqrt{X + 0.5}$ value.
Treatment means with the letter(s) in common are non-significant by DNMRT at a 5% level of significance.

Coccinellids:

The pooled data on the population of coccinellids, as shown in Table 2, reveal a significant difference between protection levels with unprotected plots showing a considerably higher coccinellid population. Specifically, the average population was 0.96 per plant in the unprotected plots, compared to 0.54 per plant in the protected plots. The significant difference between these protection levels indicates that the unprotected plots, which were not subjected to insecticide applications, provided a more favourable environment for coccinellids, allowing their population to thrive. When examining the effect of different Bt cotton hybrids on coccinellid populations, no statistically significant difference was observed. This suggests that the differences in predator populations were not influenced by the specific hybrid cultivated but were more related to the pest management regime in place. Additionally, no significant interaction was found between the hybrids and protection levels, meaning that the response of the coccinellid population to the protection levels remains consistent across all hybrids. Although the statistical analysis revealed non-significant differences between hybrids, there were some observable trends in the data. Higher coccinellid activity was recorded in hybrids such as G. Cot. Hy.10 BG II, ATM BG II, and Ajeet 155 BG II, and these were comparable to the levels observed in RCH 2 BG II. While these trends were not statistically significant, they may indicate that certain hybrids could potentially provide more favourable conditions for natural enemies like coccinellids. However, the lack of significance in these results emphasizes that any variation in coccinellid activity is most likely driven by the management practices (i.e., protected vs. unprotected conditions) rather than inherent differences between the hybrids themselves.

Table 2: Impact of hybrids and protection on population of coccinellids in cotton (Pooled)

Treatments	No. of coccinellids/plant				
Main\ Sub plot	H_1 (ATM BGII)	H_2 (Ajeet 155 BGII)	H_3 (RCH 2 BGII)	H_4 (G.Cot.Hy. 10 BGII)	Mean
P_1 (Protected)	1.02 (0.54)	1.02 (0.54)	1.01 (0.52)	1.04 (0.58)	1.02^b (0.54)
P_2 (Unprotected)	1.21 (0.96)	1.21 (0.96)	1.20 (0.94)	1.23 (1.01)	1.21^a (0.96)
Mean	1.12 (0.75)	1.12 (0.75)	1.10 (0.71)	1.13 (0.78)	1.11 (0.73)
Interactions	Protection (P)	Hybrid (H)	P x H	C.V. (%) Main	
S. Em. ±	0.04	0.009	0.01	7.60	
C. D. at 5%	0.13	NS	NS		
	Period (Y)	P x Y	H x Y	P x H x Y	C.V. (%) Sub
S. Em. ±	0.01	0.02	0.02	0.03	6.92
C. D. at 5%	0.04	0.06	NS	NS	

Notes:
Figures in parentheses are retransformed values, those outside are $\sqrt{X+0.5}$ value
Treatment means with the letter(s) in common are non-significant by DNMRT at 5% level of significance

Spiders:

The data on spider populations was outlined in Table.3, show a significant difference between the protection levels in Bt cotton fields. Specifically, the unprotected plots recorded a notably higher spider population, averaging 0.94 spiders per plant, compared to the protected plots, which had a significantly lower average of 0.54 spiders per plant. This significant difference suggests that the absence of chemical interventions in the unprotected plots favoured the survival and activity of spiders, allowing them to maintain a higher population. In contrast, the use of insecticides in the protected plots appeared to have a suppressive effect on spider populations. The variation in spider population was primarily driven by the protection levels, with the population differing based on whether the plots were protected or unprotected. However, when examining the effect of different Bt cotton hybrids, the results indicated that the hybrids themselves did not have a significant influence on spider population levels. In other words, the presence of spiders was consistent across different hybrids, and the variations observed were largely dependent on the management practices rather than the specific Bt cotton varieties being grown. Among the hybrids, G. Cot.Hy.10 BG II, ATM BG II, and Ajeet 155 BG II recorded slightly higher spider populations, ranging from 0.73 to 0.78 spiders per plant, and these values were comparable to the population observed in RCH 2 BG II, which averaged 0.71 spiders per plant. While these hybrids showed marginally higher activity of spiders, the differences were not statistically significant. This indicates that while certain hybrids may offer marginally better conditions for spider populations, these differences are not pronounced enough to be considered significant. The interaction between protection levels and hybrids in relation to spider population was also

found to be non-significant. This means that the effect of the protection level on spider populations was consistent across all hybrids, and there was no specific hybrid that responded differently to the protection regime. The lack of significant interaction further reinforces the conclusion that the primary driver of spider population dynamics is the level of pest management employed, rather than the genetic characteristics of the *Bt* hybrids themselves.

Table.3: Impact of hybrids and protection on population of spiders in cotton (Pooled)

Treatments	No. of spiders/plant				
Main\ Sub plot	H_1 (ATM BGII)	H_2 (Ajeet 155 BGII)	H_3 (RCH 2 BGII)	H_4 (G.Cot.Hy. 10 BGII)	Mean
P_1 (Protected)	1.02 (0.54)	1.03 (0.56)	1.01 (0.52)	1.04 (0.58)	1.02^{b} (0.54)
P_2 (Unprotected)	1.19 (0.92)	1.21 (0.96)	1.19 (0.92)	1.23 (1.01)	1.20^{a} (0.94)
Mean	1.11^{ab} (0.73)	1.12^{ab} (0.75)	1.10^{b} (0.71)	1.13^{a} (0.78)	1.11 (0.73)
Interactions	Protection (P)	Hybrid (H)	P x H	C.V. (%) Main	
S. Em. ±	0.03	0.008	0.01	4.65	
C. D. at 5%	0.10	0.02	NS		
	Period (Y)	P x Y	H x Y	P x H x Y	C.V. (%) Sub
S. Em. ±	0.009	0.01	0.02	0.03	6.16
C. D. at 5%	0.03	0.03	NS	NS	

Notes:

Figures in parentheses are retransformed values, those outside are $\sqrt{x+0.5}$ value

Treatment means with the letter(s) in common are non-significant by DNMRT at 5% level of significance

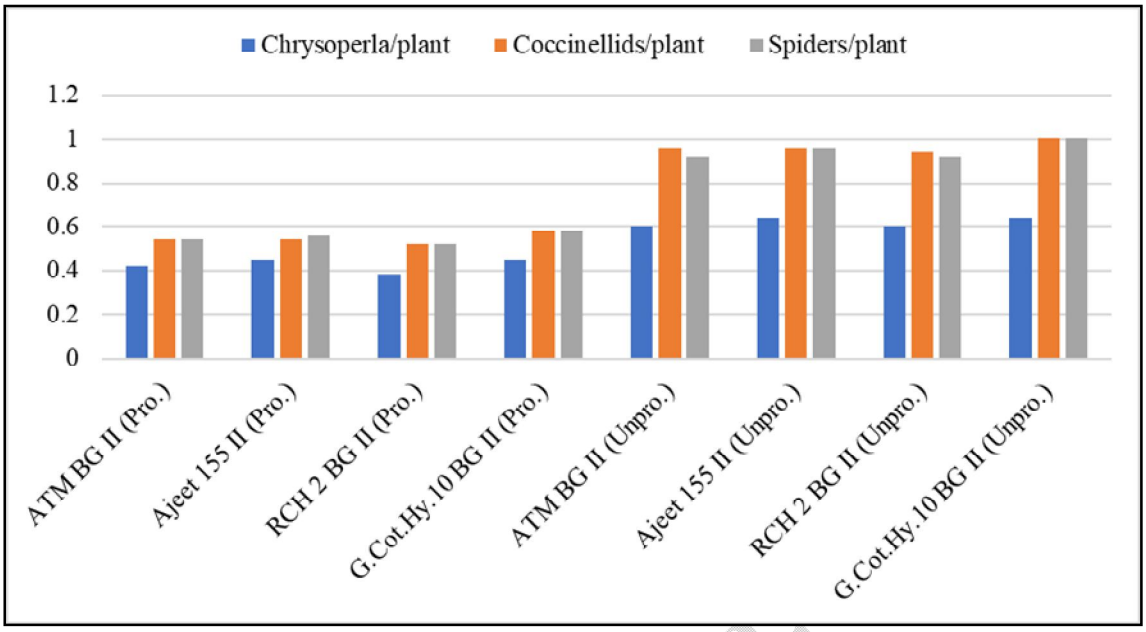


Fig.1: Natural enemies in *Bt* cotton under protected and unprotected condition

The population of natural enemies observed from 35th SW which is similar to findings of [6] who reported natural enemies (coccinellids and spiders) was noticed from 1st week of September (35th SW) to 1st week of November (44th SW) with higher activity during the 38th SW, there is no any difference in predator population under protected and unprotected condition which is similar to result of [7] who reported that the beneficial insects *viz.*, coccinellids, green lacewing, hover fly and big eyed bug were not so severely affected in the treated and control fields and no statistical differences noted in their densities. The ladybird beetle population became active in the 5th week after sowing (WAS), corresponding to the second week of August (0.14 adults per plant). Its numbers steadily increased, reaching a peak of 2.94 adults per plant by the 13th WAS, which was the first week of October [8]. The green lacewing population became noticeable at the 11th week after sowing (WAS), corresponding to the third week of September (0.20 adults per plant). Its population gradually increased and reached a peak of 1.73 adults per plant during the second half of October (16th WAS) [9]. The spider population first appeared during the 5th week after sowing (WAS), in the second week of August (0.38 spiders per plant). Its numbers quickly increased, peaking at 12.72 spiders per plant by the 18th WAS, in the first half of November. Afterward, the population declined but remained active until the end of the crop season, with 3.24 spiders per plant [10]. The populations of predators, including Coccinellids and Chrysopa, between the 30th and 50th Standard Meteorological Week (SMW) [11]. The average population of Coccinellids, Chrysoperla, and syrphids was recorded as 0.89, 0.78, and 1.00 per plant on RCH 2 Bt cotton, and 0.91, 0.75, and 1.04 per plant on RCH 2 non-Bt cotton, respectively [12]. According to [13] that *C. cornea* populations on cotton began increasing in early August, peaking at 4.11 per 15 bags by September 16. The population of coccinellid beetles started on September 16, rising to a peak of 2.78 on October 16, and 1.44 on November 2 per

15 bags. The population of natural enemies was higher in protected conditions compared to unprotected conditions [14].

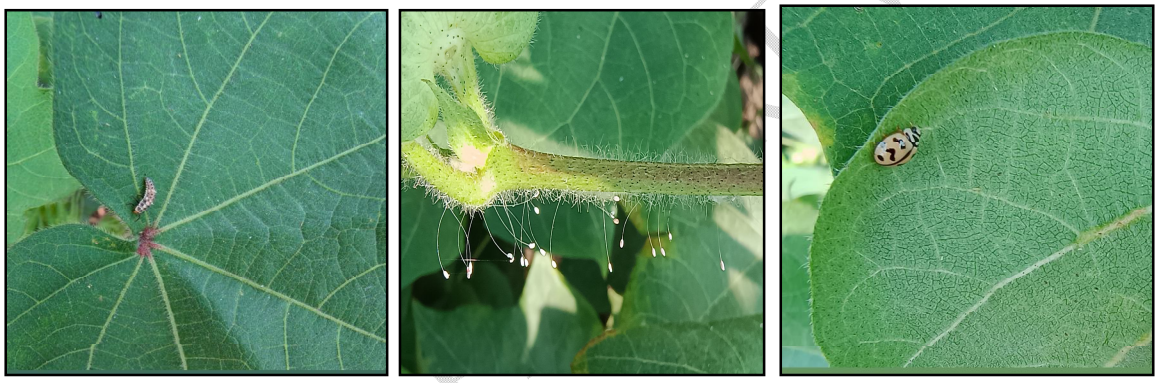
Conclusion:

Unprotected plot recorded significantly higher *Chrysoperla* larval population (0.62/plant) than protected plot (0.42/plant). There was no any significant difference found among the different hybrids. The coccinellids population recorded higher under unprotected plot (0.96/plant) than the protected plot (0.54/plant). However, the coccinellids higher activity recorded in G. Cot. Hy.10 BG II, ATM BG II and Ajeet 155 BG II and was comparable to RCH 2 BG II. Significantly higher spiders' population was recorded under unprotected plot (0.94/plant) as compared to protected plot (0.54/plant). Among the hybrids G. Cot. Hy.10 BG II, ATM BG II and Ajeet 155 BG II recorded higher spiders population ranged from 0.73 to 0.78/plant and comparable to RCH 2 BG II (0.71/plant).

Reference

1. Barwale RB, Gadwal VR, Zehr U, Zehr, B. Prospects for Bt cotton technology in India. 2004.
2. Rawal R, Dahiya KK, Lal R, Kumar A. Population dynamics of natural enemies on bt/non bt cotton and their correlation with weather parameters. *Journal of Applied and Natural Science*. 2017; *9*(4), 2360-2365.
3. Phulse VB, Udikeri SS. Sucking pests and predator dynamics in Bt and non Bt cottons grown in traditional and non-traditional locations. *Journal of Cotton Research and Development*. 2017; *31*(2), 309-316.
4. Luttrell RG, Teague TG, Brewer MJ. Cotton insect pest management. In: Cotton, 2nd ed., Agronomy Monograph 57. American Society of Agronomy, Inc., Crop Science Society of America, Inc., Soil Science Society of America, Inc., Madison, WI, 2015. pp. 509–546. https://doi.org/10.2134/ agronmonogr57.2014.0072.
5. Simon-Delso N, Amaral-Rogers V, Belzunces LP, Bonmatin JM, Chagnon M, Downs C, Goulson D. Systemic insecticides (neonicotinoids and fipronil): trends, uses, mode of action and metabolites. Environ. Sci. Pollut. Res., 2015; 22: 5-34. https://doi.org/10.1007/s11356-0143470-y
6. Chavan DR, Zanwar PR, Ramesh KB, Babu HS, Manjunatha MK. Population dynamics of sucking pests and their natural enemies of Bt cotton. Adv. Life Sci. 2016; 5(3): 954-958.
7. Sarwar M, Sattar M. An Analysis of comparative efficacies of various insecticides on the densities of important insect pests and the natural enemies of cotton, *Gossypium hirsutum* L. Pak. J. Zool. 2016; 48(1): 131-136.
8. Chauhan RP, Vekaria MV, Chaudhary HK, Chaudhary NJ. Seasonal incidence of sucking pests and their natural enemies in Bt cotton. Journal of Entomology and Zoology Studies, 2017; 5(5), 1274-1282.
9. Nemade PW, Budhvat KP, Wadaskar PS, Patil BR. Status of leaf hopper (*Amrasca biguttula biguttula* Ishida) in Bt cotton and impact of weather parameters and natural enemies on its population. Journal of Cotton Research and Development. 2015; 29(2):287-292.

10. Bhute NK, Bhosle BB, Bhede BV, More DG. Population dynamics of major sucking pests of Bt cotton. Indian Journal of Entomology. 2012; 74(3):246-252.
11. Parsai SK, Shastry PP. Seasonal incidence of key pests and their natural enemies on cotton. National Symposium on "Bt-cotton: Opportunities and Prospects" at CICR, Nagpur. 2009; 125-126.
12. Udikeri SS, Patil BV, Basavanagoud K, Khadi BM, Kulkarni KA, Vamadevaiah HM. Impact of Bt transgenic cotton on population dynamics of aphids and natural enemies. Indian J. of Agril. Sci. 2012; 82 (6): 555–560.
13. Arshad M, Khan HAA, Rehman MA, Saeed NA. Incidence of insect predators and parasitoids on transgenic Bt cotton in comparison to non Bt cotton varieties. Paki. J. Zool., (2015; 47 (3): 823-829.
14. Buttar GS, Kumar R, Jat SL. Comparative performance of cotton subjected to organic and synthetic fertilizer under protected and unprotected conditions of insects. *Journal of Entomological Research*. 2010; *34*(2), 135-137.



(a) Chrysoperla larvae (b) Chrysoperla egg (c) Coccinellids

(d) *Eriovixia excelsa*

Plate 1 : Microphotographs of Natural Enemies in different Bt Cotton hybrids