



# Evaluating Insecticide Spray Regimes to Manage Cotton Leafhopper, *Amrasca devastans* (Distant): Their Impact on Natural Enemies, Yield and Fiber Characteristics of Transgenic *Bt* Cotton

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## ABSTRACT

Cotton leafhopper *Amrasca devastans* (Dist.) is a major sucking insect pest of cotton and unfortunately natural enemies are not effective for its control. Insecticides are the only option for the management of this pest in Pakistan. On the introduction of transgenic cotton, insecticides are applied to manage sucking insect pests only. In the present study, nine combinations of conventional and new insecticides were evaluated against *A. devastans* and natural enemies for two years in transgenic cotton field. In every spray regime three insecticides were applied after an interval of 15 days. Spray regime of dimethoate, chlorfenapyr and acephate was the best in reducing *A. devastans* populations. However, the efficacy of insecticides against *A. devastans* was reduced when insecticides having same mode of action were applied in a spray regime. All the regimes proved toxic to varying degree to generalist predators like *Orius* spp., *Geocoris* spp., *Chrysoperla carnea* (Stephens), *Coccinellids* spp. and spiders. Spray regimes differed in their impact on growth and reproductive parameters of *Bt* cotton, which was directly related to *A. devastans* infestation. Overall higher root length (cm), shoot length (cm), number of leaves, yield (kg ha<sup>-1</sup>), ginning out turn (GOT %), micronaire (µg inch<sup>-1</sup>), staple length (mm) and fiber strength were found in plants treated with spray regime. These results will help to manage *A. devastans* on transgenic cotton and will ultimately reduce the yield and fiber quality losses.

## Article Information

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## Authors' Contributions

RS conducted the study and collected data. RS and M Razaq analyzed the data and wrote the article. M Razaq, M Rafiq and MN supervised the work.

## Key words

Chemical control, toxicity, insecticide rotation, organophosphates, predators, transgenic, *Bt* cotton, cotton leaf hopper, fibre characteristics of cotton spray regimes.

## INTRODUCTION

Adoption and cultivation of genetically modified crops has become a normal practice worldwide including Pakistan. More than half of the cotton grown area is occupied by cotton containing *Bacillus thuringiensis* genes in the world (known as *Bt* cotton) (Ali *et al.*, 2010; Naranjo, 2011). In 2005, *Bt* cotton was introduced in Pakistan to control insecticide resistance strains of lepidopteron pests with expected results of reduction in insecticide use (Sabir *et al.*, 2011). Due to reduced use of insecticides against cotton bollworms, the populations of sucking insect pests increased that might have been suppressed by insecticidal applications (Williams, 2006; Naranjo, 2011). In addition, transgenic *Bt* cotton attracts or enhances the growth of some sucking pest populations resulting in more insecticidal applications. Introducing transgenic varieties led to alteration in insecticidal regimes to manage non-target pests. These alterations in pesticide application regimes might affect the pest and natural enemy populations (Men *et al.*, 2004; Arshad *et*

*al.*, 2009).

Cotton leafhopper, *Amrasca devastans* (Dist.) is a principal insect pest of cotton causing more than 37% seed cotton losses. It also reduces photosynthesis activity in its hosts (Razaq *et al.*, 2014). Farmers rely solely on pesticides to manage this pest (Saeed *et al.*, 2015a), which are applied on cotton without any gap since long. Pesticides are highly effective, rapid in action, convenient to apply, usually economical and most powerful tools in pest management. However, indiscriminate, inadequate and improper use of pesticides has led to severe problems such as development of pesticide resistance, resurgence of target species, outbreaks of secondary pests, destruction of beneficial insects, as well as health hazards and environmental pollution (Yadav, 1989).

In Pakistan, resistance to pyrethroids, neonicotinoids and insect growth regulators (IGRs) has been reported in sucking insect pests of cotton such as *Bemisia tabaci* (Gennadius), *A. devastans* and *Aphis gossypii* Glover (Ahmad *et al.*, 1999; Basit *et al.*, 2011). Organophosphates like chlorpyrifos and fenitrothion have been reported to be toxic for vespidae predators of coffee leaf miner, *Leucoptera coffeella* Guérin-Ménéville (Gusmão *et al.*, 2000; Galvan *et al.*, 2002; Fernandes *et al.*, 2010). Due to continuous insecticidal applications, some natural enemies had developed resistance and

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became more effective biological control agents for pests in various cropping systems. For example, *Galendromus occidentalis* for the control of spider mites on almond and walnut in California and *Pnigalio flavipes* (Ashmead) for the control of western tentiform leafminer in Pacific Northwest (Jones *et al.*, 2009).

Previously, the efficacy and scheduling of various insecticides alone or in mixture has been documented by several authors against *A. devastans* on various crops (Razaq *et al.*, 2005; Khattak *et al.*, 2006; Shah *et al.*, 2007; Asi *et al.*, 2008; Awan and Saleem, 2012; Haq *et al.*, 2012). But there is lack of research on the efficacy of conventional insecticides in rotation with new chemistry insecticides against *A. devastans* and their impact on natural enemies on transgenic cotton. The objectives of the current study were 1) to evaluate the comparative efficacy of conventional and new chemistry insecticides in different spray regimes against *A. devastans* and their impact on natural enemies, and 2) to examine which regime improves yield and fiber characteristics of transgenic *Bt* cotton.

## MATERIALS AND METHODS

To evaluate the efficacy of different insecticidal spray regimes against *A. devastans*, transgenic *Bt* cotton (*Bt*-CIM.599) was planted on 20<sup>th</sup> May 2012 and 22<sup>nd</sup> May 2013. Plots were 10.67m × 5.34m and separated by 1.52m buffer zone. Plant to plant and row to row distance was 0.25m and 0.83m, respectively. In each year, ten treatments (n= 3 replicates each) were arranged in Randomized Complete Block Design. These treatments were nine spray regimes (including different combinations of conventional and new chemistry insecticides) and one untreated control (details of insecticides and their combinations for spray regimes are given in Tables I, II). The study was conducted under semi-arid climatic conditions at the Central Cotton Research Institute, Multan (30.12° N and 71.28° E), Pakistan. All the standard cultural practices recommended for growing cotton were followed.

The experimental field was kept unsprayed initially for the development of *A. devastans* population and its natural enemies. *A. devastans* nymphs and adults were monitored twice a week and spray was started when *A. devastans* population reached at/above economic threshold level (ETL) of one jassid per leaf (Ahmad *et al.*, 1985). Three consecutive sprays for each regime were applied at two week intervals starting from last week of June in both years. Insecticides were applied with a knapsack sprayer having a spray volume of 250 l ha<sup>-1</sup> at the pressure of three bars fitted with a hollow cone nozzle. In each year, untreated control plots were kept

unsprayed throughout the season for comparison.

Number of *A. devastans* on expended leaves (n= 30), one leaf from apical, 2<sup>nd</sup> from middle and 3<sup>rd</sup> from the bottom portion of randomly selected plant (10 plants/replicate) from each treatment were observed visually (Razaq *et al.*, 2005) after each spray at weekly interval. However, to record natural enemies whole plants were selected randomly (n=10 plants/replicate) from each treatment on each sampling date.

One week after third spray, plants (n= 3 per treatment) were removed gently from all the tested spray regimes and untreated plots. These were brought to laboratory, washed with water and spread on paper to measure root and stem length and to count number of leaves, squares and flowers.

At crop maturity, raw cotton from each plot (n= 3 plots per treatment) was picked for recording yield. Seed cotton samples (n= 100g per replicate) were packed separately in paper bags and sent to Fiber Technology Department, CCRI, Multan, Pakistan for lint testing during 2013.

### Statistical analysis

For assessing the efficacy of tested insecticides against *A. devastans* percent population reduction in different modules was calculated by using Henderson–Tilton formula (Henderson and Tilton, 1955; Kolarik and Rotrek, 2013) as follows:

$$\text{Population reduction (\%)} = \left(1 - \frac{C_a \times T_b}{C_b \times T_a}\right) \times 100$$

Where  $C_a$  is pre-treatment population in control plot;  $C_b$ , post-treatment population in control plot;  $T_a$ , pre-treatment population in treated plot;  $T_b$ , Post-treatment population in treated plot;

Data on all variables means were analysed for variance (ANOVA) by a general linear model (using GenStat Statistical Package, version 15 (VSN International, Hemel Hempstead, U.K.) which allows parametric analysis of data with normally distributed error variance without prior transformation (Batchelor *et al.*, 2006). Marginal return was calculated as the value of yield gain due to spraying, relative to the cost of spray schedule (Nabirye *et al.*, 2003). Differences between treatments means were compared using Tukey's HSD test with 5% level of probability following significant F-test.

## RESULTS

### *Amrasca devastans*

During 2012, spray started from 24<sup>th</sup> June as population crossed ETL (1 jassid leaf<sup>-1</sup>) in all plots and

**Table I.- Insecticides their groups, common names, trade names, manufacturer and dose rates applied in different regimes.**

Sr. no.	Group/ classification	Common name	Trade name	company name	<sup>a</sup> Dose (a.i.)	<sup>b</sup> Cost (100 Rs = 1 U\$)
1	Carbamate	Carbosulfan	Advantage 20%EC	FMC	200	925
2	Organochlorine	Endosulfan	Endosulfan 35% EC	FMC	280	825
3	Organophosphate	Dimethoate	Danadem Progress 40%EC	Swat Agro Chemicals	160	1,250
4	Organophosphate	Acephate	Acephate 75% SP	Jaffer Group	247	653
5	Pyrethroid	Bifenthrin	Jatara 10%EC	Jaffer Group	25	1,540
6	IGR	Pyriproxyfen	Priority 10.8 EC	KANZO Ag	54	500
7	Neonicotinoid	Imidacloprid	Confidor 20% SL	Bayer Crop Science	40	344
8	Pyrole	Chlorfenapyr	Pirate 320 SC	BASF	81	4,000
9	Thiourea	Diafenthiuron	Polo 500 SC	Syngenta	309	188

<sup>a</sup> Dose of active ingredient g or ml/ha

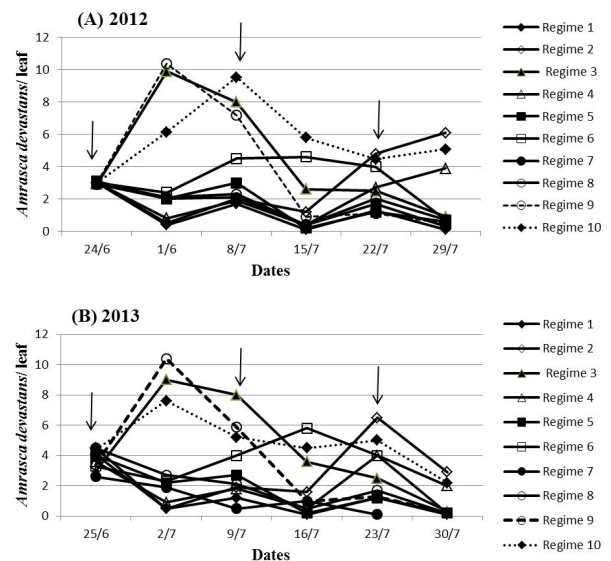
<sup>b</sup> Cost calculated/ha

**Table II.- Regime wise insecticidal treatment combinations applied in 1<sup>st</sup> 2<sup>nd</sup> and 3<sup>rd</sup> sprays.**

Spray regime no.	1 <sup>st</sup> spray	2 <sup>nd</sup> spray	3 <sup>rd</sup> spray
Regime 1	Dimethoate	Chlorfenapyr	Acephate
Regime 2	Acephate	Pyriproxyfen	Bifenthrin
Regime 3	Pyriproxyfen	Diafenthiuron	Endosulfan
Regime 4	Chlorfenapyr	Imidacloprid	Pyriproxyfen
Regime 5	Diafenthiuron	Dimethoate	Carbosulfan
Regime 6	Endosulfan	Bifenthrin	Dimethoate
Regime 7	Imidacloprid	Carbosulfan	Chlorfenapyr
Regime 8	Carbosulfan	Endosulfan	Diafenthiuron
Regime 9	Bifenthrin	Acephate	Imidacloprid

second and third sprays were applied on 8<sup>th</sup> and 22<sup>nd</sup> July, respectively. During 2013, first spray was applied on 25<sup>th</sup> June, second and third on 9<sup>th</sup> July and 23<sup>rd</sup> July in all spray regimes (Fig. 1A & B). Treatments significantly influenced *A. devastans* populations ( $F_{9,36} = 17.95, P < 0.001$ ) but the effect of year ( $F_{1,36} = 1.00, P = 0.42$ ) and treatment × year interaction ( $F_{9,36} = 0.98, P = 0.47$ ) was non-significant. Therefore, subsequent discussion is based on pooled data for two years.

On the basis of two years average, the population reduction percentage was different among all the tested regimes. After first spray, the highest average of reduction percentage (93.88%) was found in regime 1 where dimethoate, chlorfenapyr and acephate were rotated followed by regime 2 where acephate, pyriproxyfen and bifenthrin (92.52%) were applied. While negative reduction in *A. devastans* populations were observed in regime 3 (pyriproxyfen, diafenthiuron and endosulfan) and regime 9 (bifenthrin, acephate and imidacloprid) (Fig. 2A). Highest reduction percentage (90.35%) after second spray was observed in regime 1



**Fig. 1** Mean seasonal population of *Amrasca devastans* per leaf in tested spray regimes and untreated plots during study period. (A) 2012, (B) 2013. Arrows represent timing of insecticide application for each tested spray regime

(dimethoate, chlorfenapyr and acephate) and negative reduction percentages were observed in regime 2 (acephate, pyriproxyfen and bifenthrin) (-9.45%) and regime 6 (endosulfan, bifenthrin and dimethoate) (-67.79%) (Fig.2B).

After third spray, negative reduction percentages (-11.66 and -26.03%) were found in regime 2 (acephate, pyriproxyfen and bifenthrin) and regime 4 (chlorfenapyr, imidacloprid and pyriproxyfen), while highest population reduction (92.62%) was recorded in regime 1 (Fig. 2C).

On the basis of average of all three sprays maximum reduction was recorded in regime 1 (92.62%) and lowest in regime 3 (16.70%) followed by regime 9 (23.86%) (Fig. 2D).

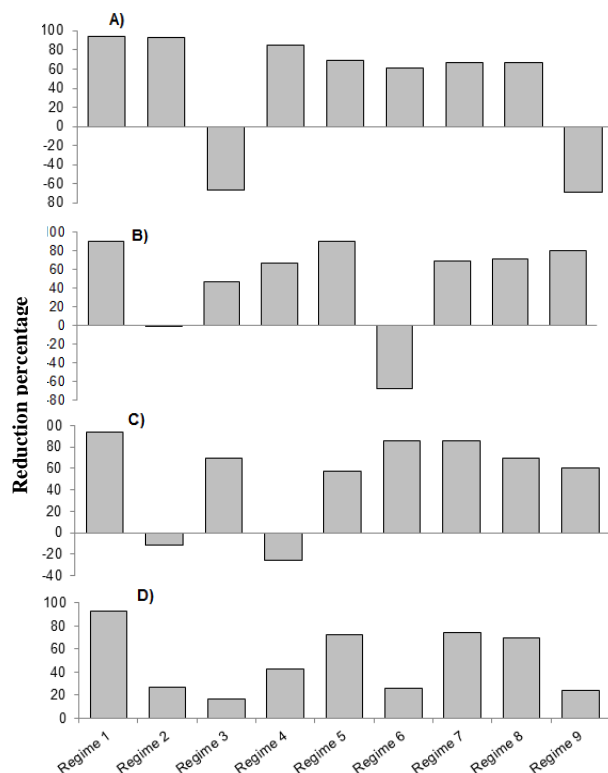


Fig. 2. *Amrasca devastans* population reduction percentages one week after spray in tested spray regimes as compared to untreated control by Henderson and Tilton formula. A) 1<sup>st</sup> spray, B) 2<sup>nd</sup> spray, C) 3<sup>rd</sup> spray D) Average of three sprays (average across two years)

#### Natural enemies

All the spray regimes significantly affected the abundance of pirate bug, *Orius* spp. ( $F_{9,36} = 3612.70$ ,  $P < 0.001$ ), big eyed bug, *Geocoris* spp. ( $F_{9,36} = 244.93$ ,  $P < 0.001$ ), green lacewing, *C. carnea* ( $F_{9,36} = 756.89$ ,  $P < 0.001$ ), lady beetle, *Coccinellid* spp. ( $F_{9,36} = 739.96$ ,  $P < 0.001$ ) and spiders ( $F_{9,36} = 1860.91$ ,  $P < 0.001$ ). More numbers of all the predators were recorded in untreated plots as compared to tested regimes plots. Toxicity of spray regimes was also consistent for almost all the taxa. Spray regime consisting of chlorfenapyr, imidacloprid and pyriproxyfen proved to be less toxic to all predators (Table III).

#### Plant growth, seed cotton yield and fiber characteristics

*Amrasca devastans* exerted profound impact on crop performance as all the plant traits [root length (cm), shoot length (cm), number of leaves, squares, flowers, yield (kg ha<sup>-1</sup>), GOT, fiber strength (tppi), micronare (μg inch<sup>-1</sup>) and staple length (mm)] were negatively and significantly correlated with *A. devastans* densities (Table IV).

Plant growth parameters and seed cotton yield of transgenic cotton significantly differed in all the treatments. Maximum root length (cm) ( $F_{9,36} = 1445.13$ ,  $P < 0.001$ ), greater shoot length (cm) ( $F_{9,36} = 392.24$ ,  $P < 0.001$ ), more number of leaves ( $F_{9,36} = 606.49$ ,  $P < 0.001$ ) and higher seed cotton yield (kg ha<sup>-1</sup>) ( $F_{9,36} = 252.49$ ,  $P < 0.001$ ) were found in regime 1 treated plants and lower in untreated control followed by regime 3 and regime 9 (Table V).

The highest yield gain percentage with maximum marginal return and profit was obtained from plots that received the spray regime 1 (Table VI). During 2013, impact of treatments on fiber characteristics was also recorded. Treatments significantly influenced fiber characteristics including GOT (%) ( $F_{9,18} = 123.64$ ,  $P < 0.001$ ), micronare (μg inch<sup>-1</sup>) ( $F_{9,18} = 72.06$ ,  $P < 0.001$ ), staple length (mm) ( $F_{9,18} = 117.73$ ,  $P < 0.001$ ) and fiber strength (tppi) ( $F_{9,18} = 118.59$ ,  $P < 0.001$ ) that were higher in regime 1 treated cotton samples as compared to all other tested regimes and untreated cotton samples (Table VII).

## DISCUSSION

In the present study, lowest mean population of *A. devastans* and its maximum reduction percentage was observed in regime 1 where insecticides with different mode of action were rotated, i.e., organophosphates (dimethoate and acephate) rotated with new insecticide (chlorfenapyr). Our results affirm the potential of dimethoate and acephate to combat *A. devastans* (Anonymous, 2012; Eijaz *et al.*, 2012; Karar *et al.*, 2013).

New chemistry insecticides were introduced in early 1990s in Pakistan. Their applications increased tremendously reaching 56% contribution among total insecticides applied on cotton in 2003 (Razaq *et al.*, 2013). The most probable reason for increased use of new chemistry insecticides is the development of resistance to conventional insecticides in insect pests of cotton (Razaq, 2006). Therefore, *A. devastans* populations might have become susceptible to conventional insecticides due their lower selection pressure seems plausible. Resistance in Pakistani populations of *B. tabaci* reverted to susceptibility to methamidophos due to their less application (Ahmad *et al.*, 2001).

**Table III.- Seasonal means of predators ( $\pm$  SE) in tested spray regimes and untreated control plots of transgenic cotton.**

Spray regimes	Predators ( $\pm$ SE)					
	<i>Orius</i> spp. (Hemiptera: Anthocoridae)	<i>Geocoris</i> spp. (Hemiptera: Lygaeidae)	<i>Chrysoperla</i> <i>carnea</i> (Neuroptera: Chrysopidae)	<i>Coccinellid</i> spp. (Coleoptera: Coccinellidae)	Spiders (Araneae)	Total predators
Regime 1	10.95 $\pm$ 0.67 d	1.93 $\pm$ 0.11 d	20.05 $\pm$ 0.54 d	3.35 $\pm$ 0.01 d	16.95 $\pm$ 0.36d	53.25 $\pm$ 1.42 d
Regime 2	5.80 $\pm$ 0.59 g	1.19 $\pm$ 0.01 f	11.30 $\pm$ 0.37 g	1.86 $\pm$ 0.07 g	10.45 $\pm$ 0.52 g	30.60 $\pm$ 1.39 g
Regime 3	12.80 $\pm$ 0.30 c	2.50 $\pm$ 0.15 c	25.55 $\pm$ 0.78 c	3.90 $\pm$ 0.15 c	19.65 $\pm$ 0.22 c	64.40 $\pm$ 1.07 c
Regime 4	14.35 $\pm$ 0.37 b	2.80 $\pm$ 0.15 b	29.49 $\pm$ 0.99 b	4.18 $\pm$ 0.04 b	22.55 $\pm$ 0.52 b	73.37 $\pm$ 1.60 b
Regime 5	4.35 $\pm$ 0.52 h	0.91 $\pm$ 0.13 g	9.34 $\pm$ 0.61 h	1.50 $\pm$ 0.15 h	8.05 $\pm$ 0.45 h	24.15 $\pm$ 1.50 h
Regime 6	3.50 $\pm$ 0.44 i	0.70 $\pm$ 0.15 g	7.45 $\pm$ 0.32 i	1.01 $\pm$ 0.13 i	5.95 $\pm$ 0.22 i	18.61 $\pm$ 1.06 i
Regime 7	11.10 $\pm$ 0.58 d	2.00 $\pm$ 0.15 d	21.05 $\pm$ 0.44 d	3.60 $\pm$ 0.04 d	17.50 $\pm$ 0.53 d	55.25 $\pm$ 1.20 d
Regime 8	7.80 $\pm$ 0.30 e	1.46 $\pm$ 0.16 e	15.70 $\pm$ 0.69 e	2.44 $\pm$ 0.14 e	14.55 $\pm$ 0.67 e	41.94 $\pm$ 1.12 e
Regime 9	6.85 $\pm$ 0.22 f	1.35 $\pm$ 0.16 ef	13.60 $\pm$ 0.40 f	2.12 $\pm$ 0.16 f	12.60 $\pm$ 0.59 f	36.52 $\pm$ 1.46 f
Untreated	19.35 $\pm$ 0.52 a	3.30 $\pm$ 0.15 a	36.06 $\pm$ 0.98 a	5.79 $\pm$ 0.16 a	31.25 $\pm$ 0.82 a	95.74 $\pm$ 2.31 a

Numbers shown are means from 10 plants per treatment and across 2012 and 2013.

Means with in a column followed by same letter are not significant (Tukey's HSD test,  $P < 0.05$ ).

**Table IV.- Correlation between *Amrasca devastans* densities and plant traits.**

Plant traits	Correlation coefficient	P value	Covariance	Standard error
Root length (cm)	-0.83	<0.001	-4.09	0.74
Shoot length (cm)	-0.81	<0.001	-12.93	2.40
Leaves	-0.80	<0.001	-16.82	3.18
Yield (kg ha <sup>-1</sup> )	-0.79	<0.001	-931.17	178
GOT (%)	-0.91	<0.001	-17.12	2.85
Fiber strength (tppsi)	-0.77	<0.001	-10.03	2.01
Micronare ( $\mu$ g inch <sup>-1</sup> )	-0.71	<0.001	-0.18	0.04
Staple length (mm)	-0.86	<0.001	-3.04	0.54

**Table V.- Transgenic cotton growth parameters ( $\pm$  SE) in tested spray regimes and an untreated control.**

Spray regimes	Growth parameters		
	Root length (cm)	Shoot length (cm)	Number of leaves
Regime 1	42.2 $\pm$ 0.44 a	71.9 $\pm$ 0.99 a	81.8 $\pm$ 0.80 a
Regime 2	24.0 $\pm$ 0.51 e	43.9 $\pm$ 0.80 f	50.8 $\pm$ 0.80 f
Regime 3	16.2 $\pm$ 1.20 hi	31.3 $\pm$ 1.00 gh	33.3 $\pm$ 1.83 hi
Regime 4	26.6 $\pm$ 0.52 e	48.0 $\pm$ 0.45 e	56.5 $\pm$ 0.68 e
Regime 5	33.2 $\pm$ 0.52 c	55.4 $\pm$ 1.06 c	67.8 $\pm$ 1.39 c
Regime 6	20.5 $\pm$ 0.43 g	41.3 $\pm$ 1.07 f	42.3 $\pm$ 0.59 g
Regime 7	34.9 $\pm$ 0.37 b	60.9 $\pm$ 2.04 b	72.8 $\pm$ 1.65 b
Regime 8	31.3 $\pm$ 0.44 d	51.9 $\pm$ 0.80 d	64.3 $\pm$ 0.66 d
Regime 9	16.8 $\pm$ 0.64 h	33.9 $\pm$ 2.00 g	35.3 $\pm$ 1.83 h
Untreated	15.0 $\pm$ 0.59 i	29.8 $\pm$ 0.44h	31.0 $\pm$ 0.85 i

Means with in a column followed by same letter are not significant (Tukey's HSD test,  $P < 0.05$ ). Data pooled across two sampling years.

**Table VII.- Fiber characteristics ( $\pm$  SE) influenced by tested spray regimes.**

Spray Regimes	GOT (%)	Micronare ( $\mu\text{g inch}^{-1}$ )	Staple length (mm)	Fiber strength (tppsi)
Regime 1	42.8 $\pm$ 0.88 a	4.10 $\pm$ 0.07 a	29.7 $\pm$ 0.35 a	103.2 $\pm$ 0.85 a
Regime 2	30.0 $\pm$ 0.53 c	3.59 $\pm$ 0.06 cde	25.0 $\pm$ 0.71 d	85.0 $\pm$ 0.71 d
Regime 3	22.5 $\pm$ 1.54 e	3.50 $\pm$ 0.07 ef	21.9 $\pm$ 0.28 e	80.0 $\pm$ 0.71 f
Regime 4	31.0 $\pm$ 0.71 c	3.61 $\pm$ 0.01 cde	26.5 $\pm$ 0.35 c	87.3 $\pm$ 0.92 cd
Regime 5	38.0 $\pm$ 0.18 b	3.70 $\pm$ 0.07 c	28.1 $\pm$ 0.19 b	91.4 $\pm$ 0.78 b
Regime 6	28.1 $\pm$ 0.60 cd	3.55 $\pm$ 0.04 def	24.3 $\pm$ 0.21 e	83.9 $\pm$ 0.71 de
Regime 7	39.0 $\pm$ 0.53 b	3.90 $\pm$ 0.04 b	28.2 $\pm$ 0.19 b	92.3 $\pm$ 0.49 b
Regime 8	37.5 $\pm$ 0.53 b	3.65 $\pm$ 0.04 cd	28.0 $\pm$ 0.14 b	90.5 $\pm$ 0.71 bc
Regime 9	25.2 $\pm$ 1.22 de	3.51 $\pm$ 0.08 ef	22.5 $\pm$ 0.14 e	81.0 $\pm$ 1.41 ef
Untreated	18.0 $\pm$ 1.32 f	3.43 $\pm$ 0.02 f	21.1 $\pm$ 0.25 e	79.0 $\pm$ 0.35 f

Means with in a column followed by same letter are not significant (Tukey's HSD test,  $P < 0.05$ ).

**Table VI.- Comparison of economic benefits among different spray regimes.**

Spray Regimes	Yield $\pm$ SE (kg ha <sup>-1</sup> )	Yield gain (%)	Marginal Return
Regime 1	3415 $\pm$ 118 a	398.5	3.9
Regime 2	1300 $\pm$ 22.3 e	89.8	0.9
Regime 3	900 $\pm$ 79.6 fg	31.3	0.3
Regime 4	1565 $\pm$ 74.2 d	128.5	1.2
Regime 5	2372 $\pm$ 114 bc	246.3	2.4
Regime 6	1150 $\pm$ 51.9 ef	67.9	0.6
Regime 7	2512 $\pm$ 111 b	266.7	2.6
Regime 8	2112 $\pm$ 155 c	208.3	2.0
Regime 9	975 $\pm$ 77.3 f	42.3	0.4
Untreated	685 $\pm$ 73.4 g	-	-

Means with in a column followed by same letter are not significant (Tukey's HSD test,  $P < 0.05$ ).

Data pooled across two sampling years; marginal returns less than 1 indicated non-profitability.

Repeated use of insecticides with same mode of action is one of the reasons for resistance development in insect pests. To minimize onset of resistance in *Helicoverpa armigera* (Hübner), use of same mode of action insecticides has been prohibited and a rotational scheme for insecticide having different modes of action has been suggested (Razaq *et al.*, 2007). However, exposing single generation of the pest with insecticides having different mode of action may develop cross resistance. Australian IRM (Insecticides Resistance Management) strategy for pyrethroids and endosulfan program by exposing one generation of pest with similar mode of action of insecticides has been proved effective in delaying resistance in pyrethroids for twelve years (Razaq *et al.*, 2013).

Our results suggest that *A. devastans* population on cotton was distinctively impacted by the type of insecticides used within each spray regime. Among the tested spray regimes, whenever pyriproxyfen or

bifenthrin was added in the regime, population flared up and results showed negative reduction percentage, reflecting their poor potential against *A. devastans*. Reduced potency of these two insecticides is also reported against *A. devastans* (Anonymous, 2011). Negative reduction percentages point out the occurrence of resurgence in insect pests (Sethi and Dilawari, 2008). Naveed *et al.* (2008) found resurgence of *B. tabaci* in bifenthrin and pyriproxyfen treated cotton plots as compared to untreated check.

The tested spray regimes against one pest may also have positive or negative impact on population of closely related pests and natural enemies (Al-Shannaf, 2010). We found that the generalist predator's community in the transgenic cotton was reduced by all the tested regimes but with varying levels. Population of *Orius* spp., *Geocoris* spp., *C. carnea*, *Coccinellids* spp. and spiders were higher in untreated control plots followed by regime 4 (chlorfenapyr, imidacloprid and pyriproxyfen) in which insecticides having novel mode of action were rotated in all three sprays. Safety of new class insecticides having novel mode of action for predators as compared to conventional insecticides is documented under laboratory and field conditions by various researchers (Nagai, 1990; Delbeke *et al.*, 1997; Elzen *et al.*, 1998; Naranjo *et al.*, 2004; Naveed *et al.*, 2008). Among the conventional insecticides, carbamates and pyrethroids are relatively more toxic to natural enemies than organophosphates (Beers *et al.*, 1993). Although new chemistry insecticides are comparatively safer for natural enemies but these chemicals with natural enemies could not reduce *A. devastans* below ETL. Resistance to these insecticides might have developed due to their continuous use (as also reported in *B. tabaci* (Basit *et al.*, 2011). Moreover, we did not measure sub lethal effects of insecticides on natural enemies which could affect their ultimate efficacy. Rotation of insecticides with same mode of



action on per generation basis can increase susceptibility of insecticides to insect pests (Razaq *et al.*, 2007). Therefore, we suggest rotational application of conventional and new chemistry insecticides to avoid resistance development and management of *A. devastans*.

Tested regimes had profound effect on plant growth and reproductive parameters. These parameters were negatively related to *A. devastans* infestation in all the regimes. Maximum root length, shoot length, number of leaves, flowers and squares were recorded in regime 1 as compared to all other regimes. Thapa *et al.* (1994) found higher efficacy of dimethoate to combat *A. biguttula biguttula* on okra with improved plant growth parameters, including taller plant, healthy pods and better quality seeds along with maximum net return as compared to other treatments. Plant growth was badly affected in untreated control plots followed by plots treated with regime 3 (pyriproxyfen→diafenthiuron→endosulfan) and regime 9 (bifenthrin→acephate→imidacloprid) treated cotton plots. Though negative population reduction percentages were found in regime 2 after 2<sup>nd</sup> and 3<sup>rd</sup> spray but growth and reproductive parameters were less affected as compared to regime 9 and regime 3. This may be due to the reason that *A. devastans* is an early season sucking pest and regime 2 treated plots received first application of acephate which suppressed population up till 2<sup>nd</sup> week and escape most vulnerable period. While regime 3 and regime 9 treated plots received first application of pyriproxyfen and bifenthrin, due to lower efficacy of these pesticides, plants undergo stress and could not be recovered by proceeding foliar applications.

Severe infestation of *A. devastans* may cause deterioration of fiber quality (Afzal and Ghani, 1953). In the present study, maximum yield kg ha<sup>-1</sup> was recorded in regime 1 as compared to all other treatments. Moreover, regime 1 also improved fiber characteristics generating higher GOT, micronaire, staple length and fiber strength as compared to other regimes and untreated control. Overall, regime 1 proved effective against *A. devastans*, ultimately increasing yield and improving fiber characteristics of transgenic *Bt* cotton. Hence, results indicated that use less effective insecticides when *A. devastans* reached to economic threshold level of one jassid per leaf may cause considerable yield loss, leading to reduced quantity and deteriorated quality of transgenic *Bt* cotton. Furthermore, supplementary laboratory studies are needed as several factors like insect density, frequency of resistant insects, age and migratory ability of insects and plant size may affect the pest mortality under field conditions (Razaq *et al.*, 2007).

In short, insecticides play a major role in the management of *A. devastans*; however the efficacy of these insecticides can be enhanced by smartly selecting

the chemistry at appropriate time keeping in view the population of natural enemies as well. Results also suggest that seed cotton yield and fiber losses can be avoided by using potential insecticides particularly in first spray and preceding insecticides rotation in different sequences to suppress *A. devastans*. Use of repeated sprays belonging to same group or having same mode of action should be avoided for resurgence management of *A. devastans* and to protect natural enemies on transgenic *Bt* cotton. The alternate option to foliar applications is seed treatment, which is also safer for natural enemies (Saeed *et al.*, 2015b).

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