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Environmental Impact of Bollworms Infestation on Cotton, *Gossypium hirsutum*

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MNA conducted the experiments, compiled and interpret the results. AF provided technical support and helped in data analysis.

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Chewing pests, Population densities, Correlation, Regression equation, Bollworms.

ABSTRACT

The present research work was conducted to evaluate the environmental changes on cotton bollworms like Pink Bollworm (PBW), Spotted Bollworm (SBW) and American bollworms (ABW) present in Multan (Southern Punjab), Pakistan. The experimental data was collected from spring 2014 to fall 2016. Eggs and adults count of these bollworms were counted throughout the cropping season. Statistix (Version 8.1) were used to analyzed the data statistically. The mean values were compared at the significance level of 5%. Bollworms (PBW, SBW, and ABW) population was lowest in April and May whereas the highest values were observed in September and October. Correlation between abiotic factor and bollworms infestation depicted non-significant negative correlation with rainfall and evening relative humidity while negative significant with minimum temperature. Linear regression equation is used to predict bollworm population to minimize economic losses.

INTRODUCTION

Cotton, wheat, rice and sugarcane are cash crops produced by farmers of an agricultural country, Pakistan. In 2015-16, more than twenty-three lacs ton cotton were produced that offer 17% of agricultural and 1.5% of national GDP (GOP, 2016). Cotton (*Gossypium hirsutum*) fiber is soft, fragile and produce in cotton bolls. Naturally cotton plant is shrubby and cultivated generally in subtropical and tropical regions. From prehistoric time cotton is used to make fiber (Huckle and Lisa, 1993). Chinese are the main fabricator for the cotton production throughout world but they totally used this for domestic purpose only. While United states are the main exporter for many years (Moulherat *et al*., 2002). In Pakistan 5 million farmers are playing their role to take the country at 5th position in production and 3rd in export of cotton (GOP, 2010). It is an important cash providing crop, which are attacked by different insect pests belongs to chewing and sucking types (Agarwal *et al*., 1981). By applying, pesticide one can control the sucking pests, but chewing pests can cause severe damage including bollworm complex named as; Spotted bollworms, *Earias insulana* (Boisduval), Pink bollworm *Pectinophora gossypiella* (Saunders); *Earias vittella* (Fabricius) and American bollworm *Helicoverpa armigera* (Hubner) (Akhtar and Farooq, 2019). These bollworms are said to be notorious insect pests which can be the reason of up to 50 % loss (Dhawan *et al*., 1998). Bollworms are called as lepidopteran, attacked different plant's fruiting bodies including cotton. They parasitized the cotton bolls and each bollworm is having different way of action on bolls, like as spotted bollworm, larvae get entry into the shoot which are the future floral bud and infestation has been observed when flower and buds are formed that results in falling of terminal shoots, squares and bolls, create a huge economic loss (Sarate *et al*., 2012; Sisterson and Tabashnik, 2004). American bollworm is polyphagous, larvae emerge from eggs after 3-4 days after that they attacked on leaves square and bolls. As for as the pink bollworms, after hatching they get entry into the bolls and completely damage the bolls inside (Reddy *et al*., 2015). Agronomic practices which are used in cotton farming enhances the probability of pest eruption, as well as ecological conditions are also playing a vital role in the population dynamics of different insect pests (Hasan *et al*., 2013; Idris and Hayat, 1997; Khaliq *et al*., 2014). A number of environmental factors are influenced on reproduction and survival of the bollworms, like as day length, humidity, rainfall and temperature. As insects are cold blooded organism, maximum and minimum temperature overwhelms the effect of environmental features (Afridi and Khan, 2015; Baloch *et al*., 1990; Reiter, 1989). Total number of eggs and Ovipositional behavior are highly influenced by the temperature (Cammel and Knight, 1992). Rainfall might have a damaging effect on the population of insect (eggs and neonate) due to dislocation or killed by rain. Metamorphosis time of insects prolonged during winter season, pigmentation changed with the change in humidity

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and temperature (Kadam and Kahaira, 1995; Schmutterer, 1990). Insects can survive only certain environmental conditions, it can be predicted the maximum activity of insects through a better understanding of environmental conditions. The present research work was undertaken to observe bollworms population and its association with abiotic factors such as maximum, minimum and average temperatures, rainfall (RF mm) and relative humidity (RH %) in cotton fields of Multan during growing to harvesting season from 2014 to 2016.

MATERIALS AND METHODS

Current research study was carried out in order to evaluate the environmental impact of bollworm infestation on squares and bolls in five different varieties of cotton during the crop season 2014-2016. Out of five varieties four were Bt (Sitara-009, MNH-988, Lalazar, IUB-33) and one Non Bt (Niab). These are the approved cotton varieties by PSC (Punjab Seed Council) Pakistan.

Experimental layout

Randomized Complete Block design was used to carry out the experiment. Five Bt and non Bt varieties were randomly assigned in three blocks of 1 Kanal (20 Marlas) plot size. Nine inch raised beds with five feet distance in each bed were managed for the seed sowing. The width of each bed in each block was 2.50 ft and length were 49.4 ft. Agronomic practices recommended by the Department of agriculture, Punjab, Pakistan were accomplished.

Field conditions

Fields were selected while keeping in mind the soil fertility status, suitable insect pressure, availability of field inputs and meteorological conditions of research area.

Seed collection

Seeds of Bt cotton were collected from cotton research institute, Multan. While Non Bt cotton seed was obtained from local seed supplier.

Seed sowing

In April of each year seed sowing was started. Squares and bolls were collected from haphazardly selected plants and bollworms infestation was noted.

Collection of data

Data about infested bolls by bollworms were noted after every ten days of interval. Percentage damaged bolls from bollworms were calculated by using following formula:

$$\text{Square Damage \%} = \frac{\text{No. of Square Damage}}{\text{Total No. of Square formed}} X\ 100$$

Following formula were used to calculate the multiple regression as well:

$$\hat{Y} = B_0 + B_1X_1 + B_2X_2 + B_3X_3 + B_4X_4 + B_5X_5$$

Meteorological data

Meteorological data was calculated from the meteorology department, Multan to correlate the weather parameters with the bollworm infestation.

Data analysis

Recorded data during the research duration was computed for ANOA (Analysis of variance by using the computer-oriented software *i.e.* Statistix (Version 8.1). The means were calculated for significant differences by LSD (least standard deviation).

RESULTS

Population dynamics of bollworms on different cotton genotypes were examined periodically.

Cotton squares and bollworm infestation

Square of different cotton varieties irrespective of Bt and non Bt, infested by Bollworms are shown in Table I.

Table I.- Bollworm infestation on square of Bt and non Bt cotton varieties.

Genotype	Bollworm infestation on squares (Standard Meteorological Week)												
	30	31	32	33	34	35	36	37	38	39	40	41	Mean
Sitara-009	0.20	0.48	0.62	0.68	0.87	1.10	1.31	1.01	1.21	3.39	3.37	3.27	1.45
MNH-988	0.70	0.71	1.13	0.95	1.28	1.37	2.83	3.10	2.42	4.21	4.61	3.42	2.22
Lalazar	0.37	0.53	0.80	1.02	1.24	1.30	2.06	3.29	2.21	4.73	4.54	3.39	2.12
IUB-33	0.39	0.88	1.31	1.25	1.60	1.60	3.17	3.39	2.75	6.10	5.60	4.52	15.18
Niab non Bt	3.33	3.53	4.11	6.88	7.55	10.44	14.66	18.53	23.31	33.2	29.87	26.76	23.68
Mean	5.83	1.22	1.59	2.15	2.5	3.16	4.8	5.86	6.38	10.32	9.59	8.27	23.68
SD	1.315	1.297	1.432	2.648	2.830	4.072	5.555	7.147	9.481	12.82	11.359	10.347	5.855
SEM (±)	0.588	0.580	0.640	1.184	1.265	1.821	2.484	3.196	4.240	5.735	5.080	4.627	

Table II.- Means (±SEM) number of adult bollworms for different periods during 2014-16.

	2014			2015			2016		
	ABW±SEM	PBW±SEM	SBW±SEM	ABW±SEM	PBW±SEM	SBW±SEM	ABW±SEM	PBW±SEM	SBW±SEM
April	0.13±0.09	0.00±0.00	0.00±0.00	0.66±0.066	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
May	1.40±0.78	0.06±0.05	0.06±0.05	12.73±4.974	10.13±4.75	2.46±1.32	0.00±0.00	0.00±0.00	0.00±0.00
June	14.00±2.19	2.80±1.61	1.93±2.17	12.66±4.126	9.66±4.08	4.06±2.40	2.53±2.11	0.80±0.73	0.86±0.79
July	15.40±3.29	14.13±4.31	7.26±3.33	12.53±4.97	11.40±3.70	3.06±1.42	6.53±3.20	3.86±2.41	4.53±2.38
August	15.66±3.82	18.93±5.67	11.80±5.39	13.20±5.34	15.20±3.76	2.93±1.47	10.13±4.83	7.06±3.70	7.46±3.33
Sept.	17.13±2.90	21.33±3.68	7.13±2.57	16.46±4.75	24.06±4.77	2.73±1.32	15.33±4.18	13.66±6.89	14.8±6.00
October	16.60±2.63	22.47±3.13	8.93±3.40	14.60±4.25	25.13±4.35	2.13±1.03	19.93±4.56	13.26±4.62	10.40±4.77

ABW, American bollworm; PBW, pink bollworm; SBW, spotted bollworm; SEM, standard error mean

Table III.- Means (±SEM) number of bollworm eggs for different periods during 2014-2016.

	2014			2015			2016		
	ABW±SEM	PBW±SEM	SBW±SEM	ABW±SEM	PBW±SEM	SBW±SEM	ABW±SEM	PBW±SEM	SBW±SEM
April	0.33±0.15	0.00±0.00	0.00±0.00	0.26±0.16	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
May	1.8±0.23	0.00±0.00	0.00±0.00	4.60±1.81	2.00±0.65	4.73±2.300	0.00±0.00	0.00±0.00	0.00±0.00
June	1.93±0.54	1.80±1.19	0.13±0.09	9.80±2.97	6.200±2.49	6.26±3.03	13.73±9.03	13.53±9.23	14.40±9.82
July	5.80±1.94	2.40±1.29	0.20±0.10	11.21±3.47	17.33±4.09	7.06±3.21	25.73±13.06	23.53±12.55	21.40±11.10
August	11.40±3.46	7.53±2.92	1.20±0.61	10.66±3.29	13.73±3.75	7.46±3.12	27.13±12.26	23.40±12.11	25.46±10.06
Sept.	14.20±4.55	15.00±3.144	0.40±0.19	8.66±2.33	12.60±2.14	7.73±2.71	16.46±5.47	24.66±11.64	32.00±11.77
October	9.46±3.12	19.26±2.55	0.26±0.15	3.12±12.8	20.46±4.07	9.33±3.83	8.66±4.49	17.53±6.24	21.80±3.10

For abbreviations, see Table II.

Bollworm infestation was started from 30th standard meteorological week (SMW) to 41 SMW *i.e.* July to October. Bt varieties shows significantly lower infestation than non-Bt. Sitara-009 shows significantly lowest infestation on 30th SMW (0.20%) and it was followed by lalazar (0.37%), IUB-33 (0.39%), MNH-988 (0.70%), while Niab non Bt showed significantly higher infestation (3.33) and it was more vulnerable for bollworm infestation. Infestation by bollworms in square was increased during 35 SMW that continue to increase till harvesting *i.e.* 41 SMW. On the basis of mean value, it was observed that minimum infestation calculated during 30th SMW and reached to maximum during 39th SMW; after that decrease was recorded.

Population count of bollworm eggs and adult

In April 2014-16 lowest population of ABW was detected and highest in October 2014-16. Population counts varied abruptly in September 2015 and reaches to its maximum. Overall it was observed that lowest and highest adult population was found in 2015 and 2016, respectively. Egg count of ABW varied through out the cropping season. It was lowest in June 2014 and highest in September 2014. In October and April 2015, July and May 2016, highest and lowest ABW counts were noted correspondingly. On the other hand, adult count of PBW showed highest and lowest number in September, April from 2014-2016 in each year (Table II). Lowest and Highest egg count of PBW were observed in April and September in each year. Adult population of SBW was found to be highest in October 2014, July 2015, September 2016 and lowest in April 2014-2016. SBW egg count showed lowest and highest figures in September, May 2014, October, April 2015 and September, April 2016 respectively in study area (Table III).

Table IV.- Correlation of Bollworms infestation with weather parameters.

Weather parameter	Correlation co-efficient (r-value)
	Damage by Bollworm
Minimum temperature (°C)	-0.739
Maximum temperature (°C)	-0.308
Evening relative humidity (%)	-0.424
Morning relative humidity (%)	0.122
Rain fall (mm)	-0.073
Wind speed (Km/h)	-0.327

Correlation with weather parameters

Environment has played a vital role in fluctuation of bollworms population. Impact of abiotic factors on bollworms infestation was computed as depicted in Table IV and Figure 1. It was observed that bollworm infestation showed significant negative correlation with minimum temperature (r=-0739), while with evening relative humidity (r=-0.424), maximum temperature (r=-0.308), with rainfall (r=-0.073) showed non-significant negative correlation, respectively. Morning relative humidity (r=0.122) exhibit positive correlation with the bollworm infestation.

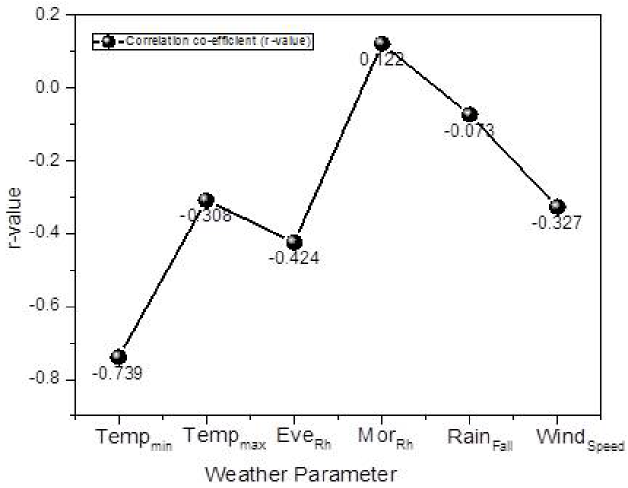

Fig. 1. Correlation coefficient of bollworms infestation with weather parameters. $Temp_{min}$, temperature minimum (°C); $Temp_{max}$, temperature maximum (°C); Eve_{Rh}, evening relative humidity (%); Mor_{Rh}, morning relative humidity (%); $Wind_{Speed}$, wind speed (Km/h).

Table V.- Multiple regression analysis between meteorological factor and bollworms infestation in squares on cotton genotypes.

	Regression	R^2
Square infested by Bollworms	Y_1=61-0.53B_1-1.45B_2-0.136B_3+ 0.147B_4+0.275B_5	0.79
	Y_2=38-1.54B_2+0.09B_4	0.73
	Y_3=36-1.25B_2	0.62

B_1, minimum temperature (°C); B_2, maximum temperature (°C); B_3, evening relative humidity (%); B_4, morning relative humidity (%); B_5, wind speed (Km/h).

Regression equation

Relationship between weather parameters and bollworms infestation were analyzed by using multiple regression equation. For this purpose, Bollworm infestation (Y) is used as dependent variable while weather parameters are independent variable as described in Table V. Changes in weather parameters and amount of change in bollworms damage clearly indicated that these factors are significantly affected *i.e.* 79% regression equation Y_1. Out of this, 73% variability accounted for evening relative humidity and minimum temperature (regression equation Y_2), while minimum temperature contributed 62% (regression equation Y_3).

DISCUSSION

Among insect's pest chewing worm can cause large financial loss to cotton crop. There are different types of bollworms, but most dangerous one is Army, Pink and Spotted bollworms. Maggots of these pests enter into cotton bolls, spoil the bolls by chewing fiber inside and became a reason of excessive loss to the crop. Insect population may fluctuate with the growing crops and with varying season. As the seed was sown, these insect moths visit field area and lay eggs. As for as crop grow, eggs hatch to larvae and get entry into bolls. Insect and egg densities vary throughout different stages of crop, which are related to moth's visit, eggs laying and hatching into worms, and then into adult stage. Environmental factors show significant variation on eggs hatching and other stages. Some difference might happen due to regional land and environment condition. In southern part of Punjab versatile climatic conditions were observed ranging from wet dry to harsh dry. It is difficult to predict weather in this area as precipitation rate is very low, some part totally depends upon rain, while in some area riverine water is supplemented with rain water.

The crops that required excess water, totally depend upon bed water for irrigation. In such harsh environmental condition only, certain insect pests can survive and flourish. The experimental site, which is center of Punjab (Pakistan), dry weather was observed round the year, winter are of quite short duration while summers are very long. In winter diurnal temperature is noticeable and minor changes occurred during summer. Proliferation of some kind of cotton chewing pests favored by such weather conditions in the study area. Pests density increases during the summer and touching to its maximum when temperature became normal. Adult of chewing Insect pests lay eggs on newly sown crop which hatch into maggots that ultimately transform into bollworms. The insect's densities and their eggs change accordingly. April or May was the month when the cotton crop was sown in each year. As seedling grew in size, eggs of these pests can be found there in the field. Obvious trend is observed when we look deeper into results. At the start of cropping season, low adult density was detected. As the more food and space for the insect available their number gradually rises. In September and/ or October but sometimes in August, highest population density was seen. Eggs count depend upon number of insects visiting crop. The difference in egg counts were observed because egg hatching take place after some days. Reduction in egg counting was due to hatching of eggs. In April of every year, bollworms i.e. PBW, SBW, and ABW had lowest and nearly same count. ABW population was found to be higher in 2014 but reduced in next two years which became higher again in October 2016. Climatic condition and pesticide spray might be the reason for this decline. In 2014 and 2015 adult ABW population enhanced abruptly, but in next months of the cropping season gradual decrease was seen. In September 2014, 2015 and 2016 highest number of PBW was observed, before and after this month their number was not significant. This picture depicted that adult number depend upon different stages of cotton plant like as appearance of flower and formation of bolls, which are extremely correlated with density of pests. SBW adults show lower count than PBW and ABW from 2014-2016. August 2014, July 2015 and September 2016 were more favored for SBW adult populations. Bollworms moth's population show fluctuation but remained active throughout the year (Glick and Graham, 1965; Qureshi *et al*., 2009; Zafar *et al*., 2013). In May- July which are the summer months decreased of bollworms were observed but become higher in October the long night month. March, April was the month where virtually no emergence of moths detected (Qureshi *et al*., 2009; Reddy *et al*., 2015). Population of larvae also show same fluctuation pattern round the cropping season. Like as pink bollworm found to be highest in August (Glick and Graham, 1965). Spotted bollworm infestation was noted during August to October (Ali *et al*., 2016; Qureshi and Ahmad, 1991) while emergence of army bollworm reached to its maximum from May to September (Ragab *et al*., 2014). Moderate and warmer areas are the best sites for the growth of Insect population than the colder regions. Precipitation rate and temperature could enhance pest populations quickly (Kavitam *et al*., 2015). Impact of temperature on development of bollworm eggs depicted that increase in temperature might disturb the development of larvae and insect pupae (Barteková and Praslička, 2006; Prasad and Bambawale, 2010; Aziz *et al*., 2011; Satti, 2012; Akram, 2013; Kumar *et al*., 2016). Environmental impact could influence on pest population and have valued correlation with each other. Pests show positive correlation with predator population while negatively correlated with temperature and rain fall (Parajulee *et al.,* 2006; Izumi *et al*., 2005; Pratheepa *et al*., 2010; Lepage *et al*., 2012; Pazhanisamy and Deshmukh, 2011; Ghosh *et al*., 2014; Pan *et al*., 2014; Kumar *et al*., 2016). Population of pink bollworm depicted negative correlation with rain fall and temperature (Khan *et al*., 2002, Wu *et al*., 2008; Tripathi, 2008; Chen *et al*., 2014; Sharma *et al*., 2016; Ali *et al*., 2016). Bollworm population had been favored by Monsoon and predators favored by winter season (Pratheepa *et al*., 2010; Hussain *et al*., 2014; Pan *et al*., 2014; Reddy *et al*., 2015). Bollworms infestation on squares was started last week of July i.e. 30th standard meteorological week in each year, and continue till the end of the cropping season. On bollworm infestation, environment played a vital role. In present research work, the infestation by bollworms is negative significant correlated with minimum temperature. Non-significant negative correlation was seen with evening relative humidity, maximum temperature and with rain fall. Same observation was reported (Kalkal *et al*., 2014; Purohit *et al*., 2006). Regarding the multiple regression analysis, it was noted that environmental factors contribute 81% variability in infestation. The prediction rate of pink and spotted bollworm population was 84% and 44% (Babu and Meghwal, 2014). That support the present study as well. The regression and correlation clearly indicated the importance of environmental factors in bollworm square infestation in cotton crop. Affective role of environmental factors was observed on the insect population round the year during cropping season. Proliferation and survival of these insect required suitable environmental conditions otherwise their survival, mortality and propagation are inevitable (Venette *et al*., 2000; Mironidis and Soultani, 2008).

CONCLUSIONS

Present study showed that Bollworms population density vary as seasonal changes occur. It became higher in summer season. Initial sprays could have some influenced but afterward rapidly increasing trend were observed. Population density reduced to lowest level by using control measures paralleling the seasonal change. Destroying eggs could be a fruitful tool to minimize the bollworm's population. Regression analysis predicted that environmental factor had significant effect on bollworm damage. Different weather parameters show negative correlation with bollworm infestation.

Statement of conflict of interest

The authors declare no conflict of interest.

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