GRANTEE: Dr. Fiaz Ahmad

JOB TITLE: Head Scientific Officer, Physiology/Chemistry Section

ORGANIZATION: Central Cotton Research Institute Multan

PROJECT TITLE: "Effect of *Bt* cotton on chemistry, microbial community structure and enzymatic activity in the rhizosphere soil"

PROJECT UPDATE: Over the last decade, the cultivation of *Bt* cotton in Pakistan has increased significantly, reaching up to more than 90% of the cotton belt. Although, the presence of the Cry1Ac protein in *Bt* cotton can provide shelter against lepidopterous species, the *Bt* toxin may have non-target effects on microbial diversity and ecosystem services. This study was conducted to evaluate the effects of *Bt* cotton on microbial populations, enzymatic activities and nutrient dynamics in the rhizosphere in comparison with non-*Bt* cotton.

In the first part of the study, extensive field surveys were conducted in three districts, specifically Multan, Lodhran and Bahawalpur of Southern Punjab. During the second part of the study, replicated field trials were conducted at four selected sites of the above districts to verify the research findings of the survey. In the survey study, rhizosphere soil samples were collected from *Bt* and non-*Bt* cotton fields in the area and analyzed for different parameters. The results revealed that the microbial population and the activity of enzymes (including dehydrogenase and phosphatase) increased slightly in the *Bt* cotton rhizosphere. Similarly, the total soil carbohydrates (CHO), active-C, soil-N (total and nitrate nitrogen), soil-P (total and extractable), soil -K, -Zn and -Fe increased in the *Bt* cotton rhizospheres.

The replicated field trials verified the findings of the survey study. Although the trend of the findings from both the studies was similar, the replicated field trials yielded more pronounced results. The *Bt* cotton rhizospheres significantly improved the population (log10 CFU g-1 soil) and activity (CO_2-C mg kg-1 day-1) of the microbes and dehydrogenase enzyme activity (µg TPF g-1 h-1) over non-*Bt* cotton rhizospheres. Moreover, the fertility related soil parameters such as OM, macro (N, P, K) and micro nutrients (Zn, Fe) were also higher in *Bt* cotton rhizospheres than the non-*Bt* cotton.

From these research findings, it is concluded that *Bt* cotton may be grown safely without having any adverse effects on soil microflora and fertility related soil parameters.

GRANTEE: Dr. Shaukat Ali

JOB TITLE: Principal Scientific Officer

ORGANIZATION: Natural Agriculture Research Centre, Pakistan Agricultural Research Council

PROJECT TITLE: "Potential risk for cross resistance development in cotton growing areas of Pakistan"

PROJECT UPDATE: The use of transgenic *Bt* cotton is tremendously dominating Pakistan's agriculture. The development of resistance to *Bt* toxins can be quite distinct, depending upon the species, selection regimen or geographical origin of the founder colony (Heckel, 1994). Information on resistance monitoring helps immensely in devising proactive resistance management strategies that can retard the rate of resistance development. A major threat to cotton farmers in Pakistan is the development of insect resistance that may be disastrous to 1.3 million cotton growers in Pakistan. Hence, regular bioassays to assess the susceptibility of the test insect to the Cry toxins will monitor the changes in the baseline that can be used in monitoring resistance that may occur due to selection pressure of the Cry1Ac toxin.

This project scrutinizes the changes in baseline toxicity, through detection of variability in the toxicity of Cry1Ac toxins to *Helicoverpa armigera* from different cotton growing regions of Pakistan during the 2013-2014 cropping season.

A research study was conducted to study growth and development of *Helicoverpa armigera* on transgenic cotton cultivars containing different levels of the Cry1Ac toxin and a non transgenic cotton cultivar. The larvae of H. armigera was collected from the chickpea fields of NARC and cotton fields of Vehari and Bakker. The insect culture was maintained and reared on an artificial diet.

Significant correlation was found between insects mortality rate and Cry1Ac toxin levels in leaves ($R^2 = 0.911$) at an exponential correlation ($y = 1.72x + 74.26$). The overall mortality rate of target pests (*H. armigera*) collected from three different regions (NARC, Vehari, Bakkar) of Pakistan ranged from 71.5-92.8% at 0.5-3.7 µg/g Cry1Ac toxins. The mortality rate of the NARC culture was found in the range of 79.5-90.0%, the Vehari culture was 71.5-88.0%, while the Bhakkar culture was 76.5-92.8%. In the case of the non-transformed control cotton cultivar, negligible larval mortality was noted. The *H. armigera* collected from the non-cotton area (NARC) and the cotton areas (Vehari and Bhakkar) showed sensitivity to a considerable level (~70%) even at 0.5 ug/g of fresh tissue weight. This reflects that *H. armigera* is still very much sensitive to Cry1Ac toxin which is currently prevailing in the *Bt* cotton of Pakistan. However, there is a need to extend the scale and spectrum of this study in the future, with other target insect pests. Continuous monitoring of field grown cotton regarding the efficacy of *Bt* toxins and the sensitivity of target insect pests will be extremely important to minimize the risk of resistance build up in the target insect pests.