

**A brief report on National Coordinated Varietal Trials (NCVT) of Cotton conducted by Pakistan Central Cotton Committee (PCCC) during the season 2018****<sup>a</sup> Muhammad Zahir Ahsan, <sup>b</sup> Hamza Afzal, <sup>c</sup> Abdul Razzaq Somro**<sup>a</sup> Scientific Officer, Cotton Research Station, Sahiwal,<sup>b</sup> MNS University of Agriculture, Multan,<sup>c</sup> Senior Cotton Breeder, ICI-Pakistan Limited, Multan.

<b>Author's Contribution</b>	M. Z. Ahsan; conducted experiments and manuscript write up, H. Rasheed; data analysis and manuscript write up, A. R. Somro; Paper write up and proof reading
<b>Article History</b>	*Corresponding email address: <a href="mailto:ahsanzahir@gmail.com">ahsanzahir@gmail.com</a> Received: 10 September 2019, Revised: 14 December 2019, Accepted: 22 December 2019, Published Online: 30 December 2019
<b>ABSTRACT</b>	Digital Object Identifier (DOI): <a href="https://doi.org/10.33865/IJCRT.001.01.0244">https://doi.org/10.33865/IJCRT.001.01.0244</a>

National Coordinated Varietal Trial was conducted to evaluate the performance of the new strains for yield potential and stability. Multi-location trial consisting of 121 strains was carried out at 13 locations under different agro-climatic zones. So different genotypes behaved differently as environment was changed hence none of the genotype was found stable in any environment. Gene expression profiling was done from four designated labs. The presence of gene and trait purity was done through event specific primers in PCR and quantities profiling was conducted by ELISA test. Most of the strains showed low purity level as well as low toxin level compared to the EPA-USA recommendation (1.5µg/g). The strains claimed as non-Bt by the respective breeder also showed the presence of Bt toxins. Hence the information is very useful for breeders as well as for other cotton stakeholders to be careful while developing high yielding varieties adoptive to multi-environment and with recommended dose of toxin level. It will also be helpful for variety registration/approval and extension department for giving general and specific recommendations of crop production.

**Key word:** Cotton varieties, fiber, Punjab, Pakistan.

**INTRODUCTION:** Cotton is crop of global importance and Pakistan is the one of the most important cotton producing as well as consuming country. Cotton is a source of fiber, cattle feed, edible oil and almost all part of the cotton plant are used extensively in several industries but in it is mainly cultivated for fiber purpose (Ahsan *et al.*, 2015) In Pakistan cotton production is important both to earn foreign exchange and for textile industry. A large number of people linked with the cotton cultivation ginning, oil industries, spinning and trade processes (Haidar *et al.*, 2016).

Since the official release of Bt cotton in Pakistan, low toxin level remained a big question mark for local varieties. The Expression of Bt gene varies according to the variety, age of the plant, part of the plant, type of the Bt gene and also its position in the genome (Adamczyk and Sumerford, 2001). The current study reports the overall yield status of strain and its adaptability across the cotton belt of Pakistan as well as Bt toxin profiling of contributed strain in National Coordinated Varietal Trial (NCVT). This method is also helpful for the breeders to make conclusion about the stability and adaptability across multiple locations.

Genotypes respond differently in different environment, that's due to their genetic makeup and environment. Some genotypes performed well in few environments and vice versa (Khan and Hassan, 2011). According to previous reports Genotypes × Environment interactions for complex traits like seed cotton yield can hinder the progress of identifying the best genotypes. G×E interaction for any cultivar reduces usefulness of genotype mean over all location for selecting and advancing superior genotypes. A genotype is considered to be stable if its variance among various environments is less. This concept of stability is very useful (Becker and Leon, 1988).

**OBJECTIVES:** Keeping in view the importance of upland cotton, the present research was planned with the aim to

study the genotype response and identify relative well adaptive and stable cultivar across different environments.

**MATERIAL AND METHODS**

**Breeding material and field testing:** A total 121 candidate strains developed by the various cotton research Institutes and private seed sector were grown on thirteen locations across cotton belt of Pakistan under National Cotton Varietal Trial during 2018-19 (table 1 to 7). Experiment was evaluated under normal growing season. Each genotype was planted in a plot having four rows of five meters length and spacing was kept 75 cm between rows and 30 cm between plants in a randomized complete block design with three replications. To maintain plant population, gap filling and thinning was carried out accordingly. All agronomic managements i.e. weeding, irrigation, inter-culturing, fertilizer application, pesticide application was done as needed. At maturity picking of plot was done and yield was calculated as kg per hectare by multiplying the yield to hectare area.

**Qualitative and quantitative profiling of Bt toxin:** All 121 genotypes were tested for Bt. gene expression profiling at four designated Labs (table 1 to 7).

1. National Institute for Genomics and Biotechnology (NIGAB) NARC Islamabad.
2. National Institute of Biotechnology and Genetic Engineering (NIBGE) Faisalabad.
3. Center of Excellence and Molecular Biology (CEMB) Lahore
4. Agriculture Biotechnology Research Institute (ABRI) AARI Faisalabad.

Approximately after eighty days of sowing confirmation and trait purity of gene was done through PCR and quantification of Cry proteins (Bt toxin) in all the entries was carried out by sandwich-ELISA. Third fully expanded leaf tissues of each entry

were used for analysis. Sandwich-ELISA was performed according to the manufacturer's instructions.

**Table 1: Coding of Strains into different sets**

Sr.	Set	Code	Total Strains	Technology Claimed
1	A	PC1801 to PC1811	11	Non-Bt
2	B	PC1812 to PC1838	27	Bt
3	C	PC1839 to PC1868	30	Bt
4	D	PC1869 to PC1895	27	Bt
5	E	PC1896 to PC18121	26	Bt

**Table 2: Non-Bt genotypes tested under Set-A.**

Codes	Genotypes	Institute	Codes	Genotypes	Institute
PC-1801	CIM-717	Central Cotton Research Institute Multan	PC-1807	CRIS-552	Central Cotton Research Institute Sakrand
PC-1802	CYTO-225	Central Cotton Research Institute Multan	PC-1808	NIAB-191	NIAB, Faisalabad
PC-1803	CRIS-613	Central Cotton Research Institute Sakrand	PC-1809	CRIS-510	Central Cotton Research Institute Sakrand
PC-1804	TH-88/11	ARI, Tandojam	PC-1810	NIAB-818	NIAB, Faisalabad
PC-1805	CIM-620	Central Cotton Research Institute Multan	PC-1811	GS-Ali-9	Gohar Seed Corporation, Multan
PC-1806	GS-Ali-7	Gohar Seed Corporation, Multan			

**Table 3: Bt genotypes tested under Set-B**

Codes	Genotypes	Institute	Codes	Genotypes	Institute
PC-1812	Bahar-07	Bahar Seed Corporation Sadiqabad	PC-1826	GH-Haadi	Cotton Research Station Ghotki
PC-1813	AA-933	Ali Akbar Seed Corporation Lahore	PC-1827	GH-Mubarak	Cotton Research Station Ghotki
PC-1814	Auriga-216	Auriga Seed Corporation Lahore	PC-1828	ICI-2121	ICI, Pakistan, Multan
PC-1815	Bahar-2017	Bahar Seed Corporation Sadiqabad	PC-1829	IUB-13	Islamia University Bahawalpur
PC-1816	BH-221	Cotton Research Station Bahawalpur	PC-1830	IR-NIBGE-11	NIBGE, Faisalabad
PC-1817	BS-18	Bandesha Seed Corporation Jahanian	PC-1831	IUB-69	Islamia University Bahawalpur
PC-1818	Bt.CIM-343	Central Cotton Research Institute Multan	PC-1832	KZ-125	Kanzo Seed Corporation Multan
PC-1819	Bt.CIM-663	Central Cotton Research Institute Multan	PC-1833	MNH-1020	Cotton Research Institute, Multan
PC-1820	CIM-602	Central Cotton Research Institute Multan	PC-1834	MNH-1026	Cotton Research Institute, Multan
PC-1821	BZU-05	Bahaudin Zakaria University, Multan	PC-1835	NIA-85	NIA, Tandojam
PC-1822	CYTO-515	Central Cotton Research Institute Multan	PC-1836	NIAB-898	NIAB, Faisalabad
PC-1823	Evyol-148	Kanzo Seed Corporation Multan	PC-1837	NS-191	Neelum Seeds Corporation, Jahanian
PC-1824	FH-444	Cotton Research Station Faisalabad	PC-1838	RH-Afnan	Cotton Research Institute, Khanpur
PC-1825	FH-490	Cotton Research Station Faisalabad			

**Table 4: Bt genotypes tested under Set-C**

Codes	Genotypes	Institute	Codes	Genotypes	Institute
PC-1839	RH-Manthar	Cotton Research Institute, Khanpur	PC-1854	Weal AG-6	Weal-Ag Seeds Corporation, Multan
PC-1840	Sahara-210	Patron Seeds Corporation, Multan	PC-1855	Badar-1 (CII)	Four Brothers Seed Corporation Multan
PC-1841	Shahab-7	Professor seed Corporation Sahiwal	PC-1856	IUB-13	Islamia University Bahawalpur
PC-1842	Shaheen-16	Aibta Seeds Corporation, Sadiqabad	PC-1857	CII-CEMB-100	CEMB, Lahore
PC-1843	Sitara-16	Agri-Farms Services, Multan	PC-1858	CII-CEMB-101	CEMB, Lahore
PC-1844	SLH-19	Cotton Research Station Sahiwal	PC-1859	NU-21 (CII)	Patron Seeds Corporation, Multan
PC-1845	SLH-6	Cotton Research Station Sahiwal	PC-1860	Sahara-2020 (CII)	Patron Seeds Corporation, Multan
PC-1846	Suncrop-5	Suncrop Seeds Corporation, Multan	PC-1861	Tahafuz-10 (CII)	Suncrop Seeds Corporation, Multan
PC-1847	CIM-602	Central Cotton Research Institute Multan	PC-1862	TJ-Max (CII)	RCA Seeds Corporation Khanewal
PC-1848	Suncrop-6	Suncrop Seeds Corporation, Multan	PC-1863	CII-Badar-2	Four Brothers Seed Corporation Multan
PC-1849	Tassco-902	Tassco Seeds Corporation TandoAllahyar	PC-1864	CII-CEMB-102	CEMB, Lahore
PC-1850	Tipu-9	V-gro SC, Sadiqabad	PC-1865	CII-ICI-2222	ICI, Pakistan, Multan
PC-1851	VH-189	Cotton Research Station Vehari	PC-1866	CII-Suncrop	Suncrop Seeds Corporation, Multan
PC-1852	VH-383	Cotton Research Station Vehari	PC-1867	CII-Tahafuz-12	Suncrop Seeds Corporation, Multan
PC-1853	Weal AG-5	Weal-Ag Seeds Corporation, Multan	PC-1868	CII-TJ-King	RCA Seeds Corporation Khanewal

**Table 5: Bt genotypes tested under Set-D**

Codes	Genotypes	Institute	Codes	Genotypes	Institute
PC-1869	CEMB-Klean Cotton-1	CEMB, Lahore	PC-1883	BS-20	Bandesha Seeds Corporation, Jahanian
PC-1870	CEMB-Klean Cotton-2	CEMB, Lahore	PC-1884	Bt. CIM-303	Central Cotton Research Institute Multan
PC-1871	Ghuri-1 (CKC)	CEMB, Lahore	PC-1885	Bt. CIM-678	Central Cotton Research Institute Multan
PC-1872	Hatf-1 (CKC)	Four Brothers Seed Corporation Multan	PC-1886	IUB-13	Islamia University Bahawalpur
PC-1873	CKC-3	CEMB, Lahore	PC-1887	Bt. CIM-789	Central Cotton Research Institute Multan
PC-1874	CKC-4	CEMB, Lahore	PC-1888	BZU-07	Bahaudin Zakaria University, Multan
PC-1875	CKC-Clean Flex	CEMB, Lahore	PC-1889	CRIS-671	Central Cotton Research Institute Sakrand
PC-1876	CKC-Hatf-3	Four Brothers Seed Corporation Multan	PC-1890	CRIS-673	Central Cotton Research Institute Sakrand
PC-1877	CIM-602	Central Cotton Research Institute Multan	PC-1891	Crystal-20	Waraich Seed Corporation, Jahanian
PC-1878	CKC-ICI-2323	Islamia University Bahawalpur	PC-1892	CYTO-510	Central Cotton Research Institute Multan
PC-1879	CKC-Sahara-Klean	Patron Seeds Corporation Multan	PC-1893	CYTO-511	Central Cotton Research Institute Multan
PC-1880	CKC-TJ-Inqilab	RCA Seeds Corporation Khanewal	PC-1894	Eagle-3	Four Brothers Seed Corporation Multan
PC-1881	BF-1	Baba-Fareed Seed Corporation, Vehari	PC-1895	Eye-111	Kanzo Seed Corporation Multan
PC-1882	BH-223	Cotton Research Station, Bahawalpur			

**Table 6: Bt genotypes tested under Set-E**

Codes	Genotypes	Institute	Codes	Genotypes	Institute
PC-1896	Eye-20	Kanzo Seed Corporation Multan	PC-18109	NIAB-1011	NIAB, Faisalabad
PC-1897	FH-155	Cotton Research Station Faisalabad	PC-18110	NIAB-135	NIAB, Faisalabad
PC-1898	FH-AM Cotton-2017	Cotton Research Station Faisalabad	PC-18111	NS-201	Neelum Seeds Corporation Jahanian
PC-1899	FH-Super Cotton-2017	Cotton Research Station Faisalabad	PC-18112	RH-670	Cotton Research Institute, Khanpur
PC-18100	GH-UHAD	Cotton Research Station Ghotki	PC-18113	IUB-13	Islamia University Bahawalpur
PC-18101	ICI-2424	Islamia University Bahawalpur	PC-18114	Rohi-1	Rohi Seeds Corporation, Rajanpur
PC-18102	IR-NIBGE-12	NIBGE, Faisalabad	PC-18115	Rustom-11	Jullundur Seeds Corporation, Rahim.Yar.Khan
PC-18103	IR-NIBGE-13	NIBGE, Faisalabad	PC-18116	SLH-33	Cotton Research Station Sahiwal
PC-18104	CIM-602	Central Cotton Research Institute Multan	PC-18117	Tassco-112	Tassco Seeds Corporation TandoAllahyar
PC-18105	IUB-71	Islamia University Bahawalpur	PC-18118	Tipu-1524	V-gro Seeds Corporation, Sadiqabad
PC-18106	Koh-i-Noor	Professor seed Corporation Sahiwal	PC-18119	VH-402	Cotton Research Station Vehari
PC-18107	MNH-1035	Cotton Research Institute, MULTan	PC-18120	Weal AG-7	Weal-Ag Seeds Corporation, Multan
PC-18108	MZM-7	Agri-Farms Services, Multan	PC-18121	Weal AG-8	Weal-Ag Seeds Corporation, Multan

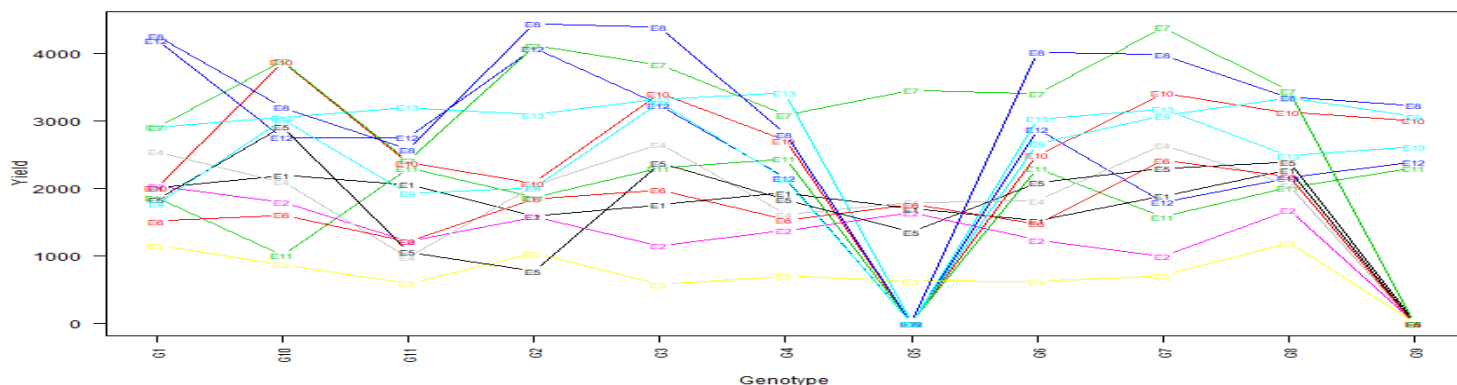
**Table 7: Locations of NCVT Sowing across different zones of Pakistan**

Sr.	Province	Zone	Station	Sets
1	Khyber Pakhtunkhawa	D.I. Khan	Cotton Research Station D.I. Khan	A,B,C,D,E
			Cotton Research Station, AARI Faisalabad	A,B,C,D,E
			Nuclear Institute for Agriculture and Biology	A,B,C,D,E
			National Institute of Biotechnology and Genetic Engineering	A,B,C,D,E
2	Punjab	Sahiwal	Cotton Research Station, Sahiwal	A,B,C,D,E
			Central Cotton Research Institute, Multan	A,B,C,D,E
			Cotton Research Station, Vehari	A,B,C,D,E
			Cotton Research Station, Bahawalpur	A,B,C,D,E
			Cotton Research Institute, Khanpur	A,B,C,D,E
			Cotton Research Station, Ghotki	A,B,C,D,E
			Cotton Research Institute, Sakrand	A,B,C,D,E
3	Sindh	Tandojan	Agriculture Research Institute, Tandojam	A,B,C,D,E
			Nuclear Institute for Agriculture, Tandojam	A,B,C,D,E
			Sindh Agriculture University Tandojam	A,B,C,D,E
			Cotton Research Station, Mirpur Khas	A,B,C,D,E
4	Balochistan	Lasbela	Cotton Research Station, Lasbela	A,B,C,D,E
		Sibbi	Cotton Research Station, Sibbi	A,B,C,D,E

**RESULTS AND DISCUSSION:** The figure 1 shows that the yield response varies in different environments. Highest yield was obtained from Genotype G7 (CRIS-552) when planted at E7 (Khanpur) i.e 4382 kg per hectare. Genotype G5 (CRIS-510) was standard at Sindh, Balochistan so did not sown in Punjab and KPK and G5 (CIM-620) was standard genotype in Punjab and

KPK so did not cultivated in Sindh and Balochistan (Ghotki, Sakrand, Tandojam, Mirpur Khas, Lasbela and Sibbi). Very low yield was observed from all genotype at E3 (Sahiwal) as compared to other locations. The genotypes G5 (CIM-620) and G9 (CRIS-510) (Standards) produced much stable yield in all environments as compared to other genotypes.

**Response Plot of Yield**



**Figure 1: Seed cotton Yield (kg per hectare) response of 11 Genotypes of Set-A at different**

The figure 2 shows response of different genotypes of Set-B at 13 cultivated locations. It was observed that highest yield was harvested from Genotype 19 (IR-NIBGE-11) only at one location i.e. E7 (Khanpur) and very low yield was obtained from Genotype G24 (NIA-85) at E8 (Ghotki) and G10 (BZU-05) from E11 (Mirpur Khas). As compared to other environments all genotypes produced low seed cotton yield when planted at Sahiwal. The response of the genotypes differed as location was changed so none of the genotypes was stable in all environments. However, genotype 24 (NIA-85) produced much stable yield except at location E8 (Ghotki).

In Set-C there were 30 genotypes the figure 3 showed their performance at all 13 locations. Maximum yield of all genotypes was harvested when they were planted at E7 (Khanpur) and E12 (Lasbela) and lowest at location E3 (Sahiwal). Highest Yield was harvested from genotypes G59 (NU-21 (CII)) and G63 (CII-Badar-2)) when planted at E12 (Lasbela) and lowest yield was obtained from genotype 45 (SLH-6) when planted at E10 (Tandojam). Very heterogeneous response of genotypes was observed with changing environment.

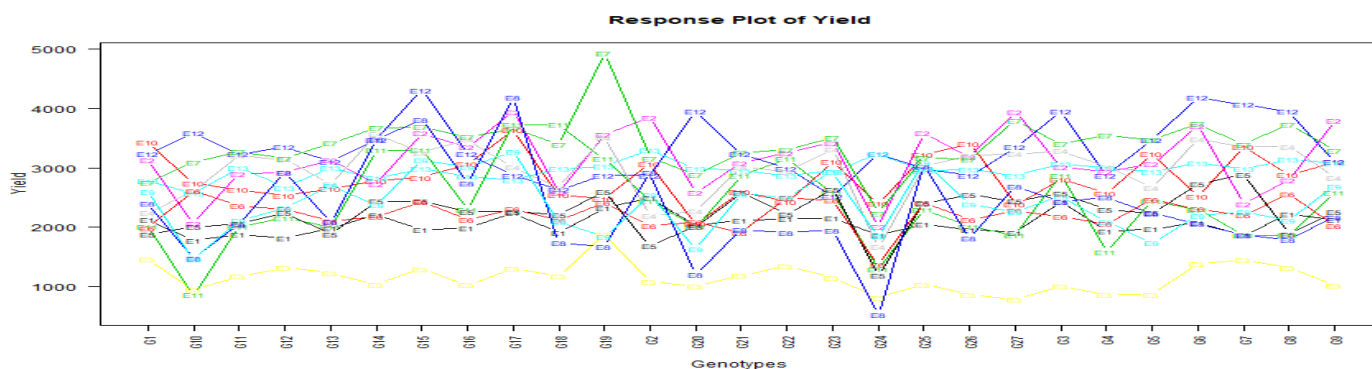


Figure 2: Yield response of 27 Genotypes of Set-B at different Locations.

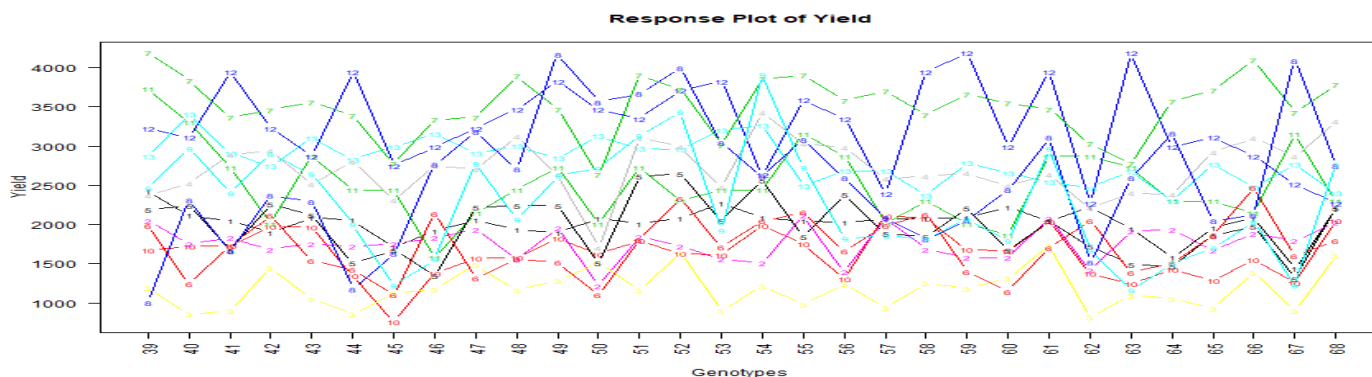


Figure 3: Yield response of 30 Genotypes of Set-C at different Locations

Yield response of 27 genotypes of set D is presented in figure 4. lowest yield was recorded from genotype 75 (CKC-Clean-Flex) Highest yield was harvested from genotype 85 (Bt.CIM-789) at E3 (Sahiwal). All genotypes produced highest yield at E7 (Khanpur) and E11 (Mirpur Khas). And (Khanpur) and lowest when planted at Sahiwal.

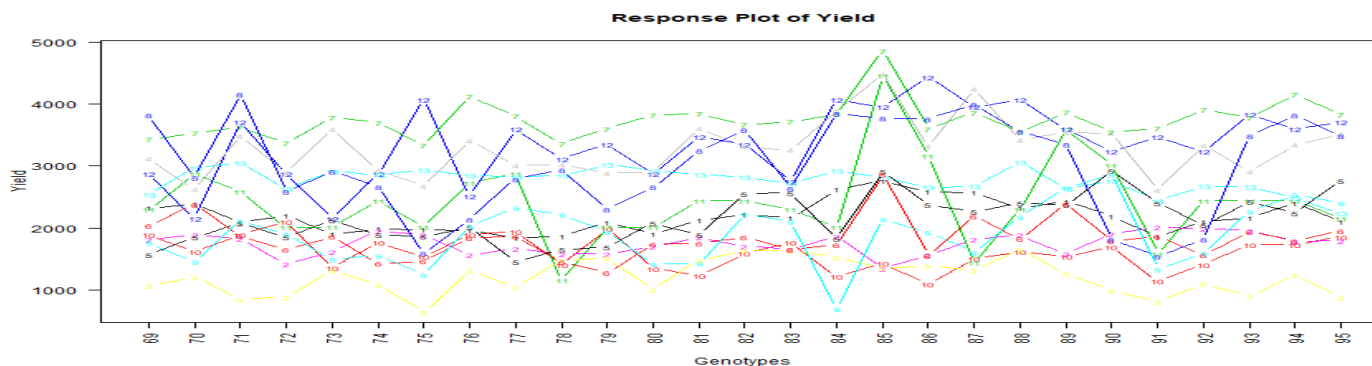


Figure 4 Yield response of 27 Genotypes of Set-D at 13 different Locations.

Yield response of 26 genotypes of set E is presented in figure 5. 108 (MZM-7) at E9 (Sakrand), 105 (IUB-71) at E10 (Tandojam) Highest yield was harvested from genotypes 98 (FH-AM-Cotton-2017), 100 (GH-Uhad) and 115 (Rustom-11) when highest yield at E8 (Ghotki) and E12 (Lasbela) and lowest when planted at E8 (Ghotki). Lowest yield was produced by genotype planted at E3 (Sahiwal).

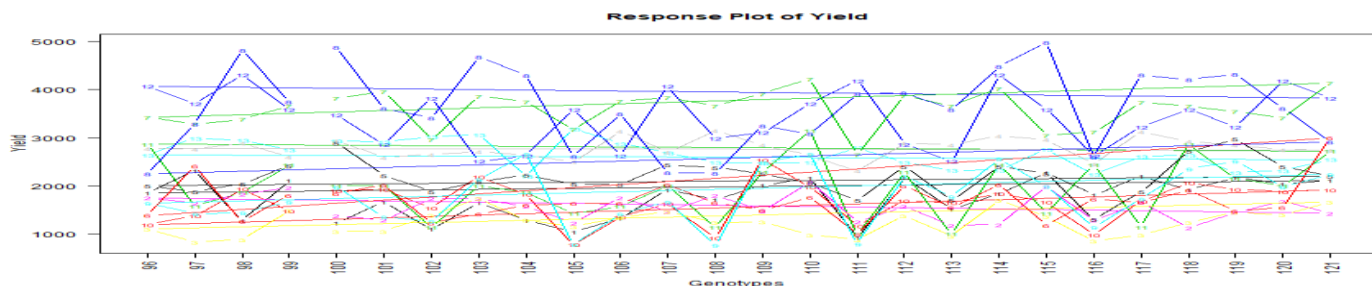


Figure 5: Yield response of 26 Genotypes of Set-E at 13 different Locations.



Polygon view of biplot What-won-where is essential for determining the possible existence of mega environment in the region (Yan, 2001; Rad *et al.*, 2013). By connecting the genotypes, a polygon is formed and rays perpendicular to the sides of the polygon divided the polygon into four sectors (Figure 6).

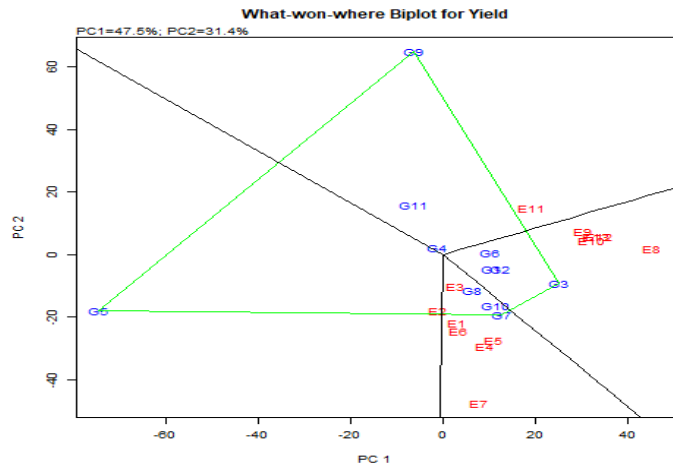


Figure 6: Polygon view of GGE Biplot based on environment scaling for What-Won-Where pattern of genotypes of Set-A and environments.

These eleven genotypes were distributed among four sectors but environments fell only in three sectors. Each sector indicated the interaction of genotype with specific environment. Genotypes G3 (CRIS-613), G9 (CRIS-510), G7 (CRIS-552) and G5 (CIM-620) were forming the polygon and suggesting the high adaptability in their respective environment. Genotype G9 (CRIS-510) was best performing in environment 11 (Mirpur Khas) however, genotypes G3 (CRIS-613), G2 (CYTO-225) and G6 (GS-Ali-7) performed better at E8, E9, E10, E12, E13 and E14 (Ghotki, Sakrand, Tandojam, Lasbela, and Sibbi). Genotype G7 (CRIS-510), G8 (NAIB-191) and G10 (NAIB-818) performed best at E1, E3, E4, E5, E6 and E7 (D.I. Khan, Sahiwal, Multan, Vehari, Bahawalpur, Khanpur). Genotype G5 (CIM-620) resided in sector where no environment was evident it indicated that this genotype did not perform well in any environment.

GGE biplot divided twenty-seven genotypes among three sectors but environments fell only in two sectors. Each sector indicated the interaction of genotype with specific environment. Genotypes G17 (ICI-2121), G19 (IR-NIBGE-11) and G24 (NIA-85) were present on the corner of the polygon and suggesting the high adaptability in their respective environments. Genotype G20 (IUB-69) along with G1 (Bahar-07), G4 (Bahar-2017), G5 (BH-221), G10 (BZU-05), G11 (CYTO-515), G13 (FH-444) and G20 (IUB-69) were present in a sector where no environment was present so these genotypes were considered non responsive to any environment. G 19 (IR-NIBGE-11) along with G3 (Auriga-216) and G6 (BS-18) were highly adoptive to the environment E 7 (Khanpur), E4 (Multan), E5 (Vehari) and E12 (Lasbela) whereas G17 (ICI-2121), G2 (AA-933), G15 (GH-Haadi), G25 (NIAB-898), G16 (GH-Mubarak), and G9 (CIM-602) performed better in environments E8 (Ghotki), E9 (Sakrand), E10 (Tandojam), E11 (Mirpur Khas) and E2 (Faisalabad), however genotypes G3 (CRIS-613), G2 (CYTO-225) and G6 (GS-Ali-7) better performed at E8, E9, E10, E12, E13 and E14 (Ghotki, Sakrand, Tandojam, Lasbela, and Sibbi). Genotype G7

(CRIS-510), G8 (NAIB-191) and G10 (NAIB-818) performed best at E1, E3, E4, E5, E6 and E7 (D.I. Khan and Sahiwal)(Figure 7)

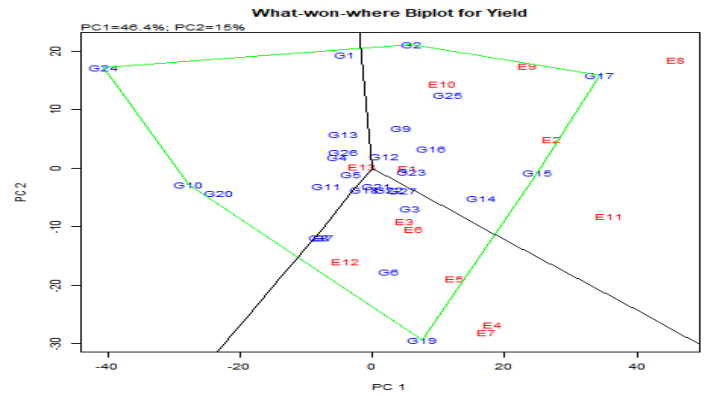


Figure 1: Polygon view of GGE Biplot based on environment scaling for What-Won-Where pattern of genotypes of Set-B and environments.

There were thirty genotypes in Set-C and they formed 7 corner polygon. Genotype 54 (Weal-ag-6), G39 (RH-Manthar), G45 (SLH-6), G67 (CII-Tahafuz-12), G49 (Tassco-902) and G52 (VH-383) were on the corner of the polygon suggesting that they were highly adoptive to the respective environment. Polygon was divided into seven sectors however four sectors don't have any environment the varieties present in these sectors did not show adoptive to any environment. Similarly environment E8 (Ghotki) and E12 (Lasbela) were present in a sector where no genotype was present suggesting that none of the genotypes performed well in these environments. Genotypes 52 (VH-383) G51 (VH-189), G61 (CII-Tahafuz-10), G55 (Badar-1 (CII) and G47 (CIM-602) performed ideally in E3 (Sahiwal) and E13 (Sibbi). Genotype 54 (Weal-ag-6), G68 (CII-tj-King), G42 (Shaheen-16) and G40 (Sahara-210) appeared better adoptive to the environment E9 (Sakrand), E5 (Vehari), E7 (Khanpur) and E6 (Bahawalpur). Most of the genotypes present in Set-C were not adoptive to any environment under test (figure 8)

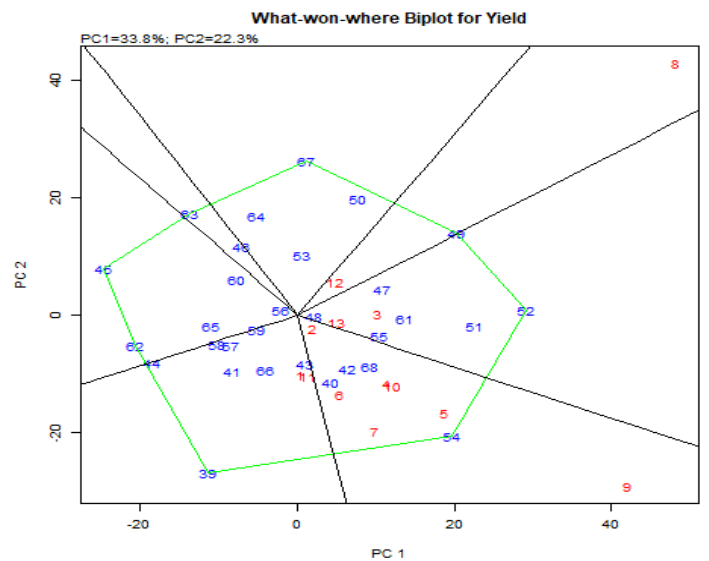


Figure 8: Polygon view of GGE Biplot based on environment scaling for What-Won-Where pattern of genotypes of Set-C and environments.

Twenty-seven genotypes in Set-D formed five corner polygon which is divided into five sectors. Genotypes G70 (CEMB-Clean-Cotton-2), G84 (Bt. CIM-303), G85 (Bt. CIM-678), G90 (CRIS-673) and G91 (Crystal-20) were at the corner of the polygon showing adaptability to all the environments. G91 (Crystal-20) along with G75 (CKC-Clean-Flex), G79 (CKC-Sahara-Klean) and G92 (CYTO-510) were in a sector where no environment was present so these genotypes did not show adoptive to any tested environment. Genotypes G84 (Bt. CIM-303), G87 (Bt. CIM-789), G88 (BZU-07), G71 (Ghauri-1 CKC), G81 (BF-1) and G69 (CEMB-Clean-Cotton-1) were the best performer genotypes in environment E8 (Ghotki) and E12 (Lasbela). The genotypes well adopted to the environments E11 (Mirpur Khas), E9 (Sakrand), E5 (Vehari), E6 (Bahawalpur) and E7 (Khanpur) were G85 (Bt. CIM-678), G89 (CRIS-671), G86 (IUB-13) and G94 (Eagle-3). Genotypes G90 (CRIS-673), G76 (CKC-Hatf-3), G70 (CEMB-Clean-Cotton-2), G77 (CIM-602) and G83 (BS-20) performed excellent at E10 (Tandojam) (figure 9).

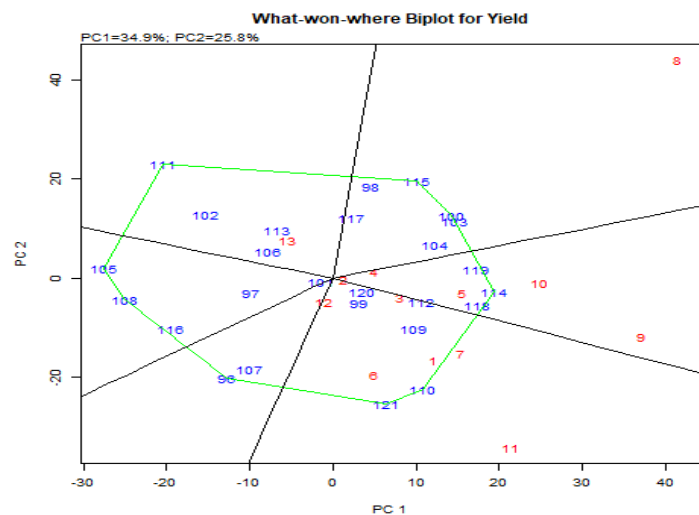


Figure 9: Polygon view of GGE Biplot based on environment scaling for What-Won-Where pattern of genotypes of Set-D and environments.

There were twenty-six genotypes in Set-E. GGE biplot divided these genotypes into six sectors. The genotypes G111 (NS-201), G115 (Rustom-11), G114 (Rohi-1), G121 (Weal-ag-8), G96 (Eye-20) and G105 (IUB-71) were at the corner of the polygon. There was no environment in three sectors suggested that the genotypes present in these sectors are not adoptive to any tested environment. Genotypes G111 (NS-201), G102 (IR-NIBGE-12), G103 (IR-NIBGE-13) and G106 (Koh-i-Noor) performed well at E13 (Sibbi) Genotypes G121 (Weal-ag-8), G110 (NIAB-135), G109 (NIAB-1011) and G99 (FH-Super-Cotton-2017) were well adopted to E11 (Mirpur Khas), E1 (D.I. Khan), E6 (Bahawalpur) and E7 (Khanpur) in a same way genotypes G114 (Rohi-1) G118 (Tipu-1524) and G119 (VH-402) were adopted to environment E9 (Sakrand), E10 (Tandojam)

and E5 (Vehari). In Set-E most of the genotypes were not adoptive to any environment under test.

**Cry1ac Gene (Event MON-531) Bollgard-I Technology:** The genetic transformation event (MON-531) and responsible gene for cry protein was investigated using PCR which successfully verified that all 121 candidate cotton strains had MON-531 event except CRIS-552 which was claimed as non-Bt strains. This event has Cry1ac gene of soil born bacterium i.e. *Bacillus thuringiensis*. Trait purity was in a range of 26% to 100%. The expression level of Cry1ac was measured in a range of 0.79 µg/g to 3.56 µg/g fresh leaf weight, most of the varieties exhibited low toxin expression as compared to the EPA-USA recommended toxin dose (1.5 µg/g). Strains CII-Tahafuz-12, CII-CEMB-101, TH-88/11 gave the highest expression level of Cry1ac (3.56, 2.88 and 2.84 µg/g, respectively). Eleven varieties were claimed as non-Bt but all lab test showed the presence of cry1ac (Event MON-531) however trait purity and expression level was very low.

**RR/GTG gene (event MON-1445) bollgard-III technology:** Eleven strains were claimed as Bollgard III technology having event MON-1445. PCR confirmed the presence of this event in all strains, trait purity was 6.67% (CKC-ICI-2323) to 100% (CKC-4 and CKC-Hatf-3). However, ELISA test showed variable amount of protein i.e. 0µg/g in CEMB-Kean Cotton -1 to 1.23µg/g in CKC-3.

## References

- Adamczyk, J. J. and D. Sumerford, 2001. Potential factors impacting season-long expression of cry1ac in 13 commercial varieties of bollgard® cotton. *Journal of Insect Science*, 1(1): 1-6.
- Ahsan, M. Z., M. S. Majidano, H. Bhutto, A. W. Soomro, F. H. Panhwar, A. R. Channa and K. B. Sial, 2015. Genetic variability, coefficient of variance, heritability and genetic advance of some *Gossypium Hirsutum* L. Accessions. *Journal of Agricultural Science*, 7(2): 147-151.
- Becker, H. and J. Leon, 1988. Stability analysis in plant breeding. *Plant breeding*, 101(1): 1-23.
- Haidar, S., M. Aslam and M. A. U. Haq, 2016. Niab-852: A new high yielding and better fibre quality cotton mutant developed through pollen irradiation technique. *Pakistan Journal of Botany*, 48(6): 2297-2305.
- Khan, N. and G. Hassan, 2011. Genetic effects on morphological and yield traits in cotton (*Gossypium Hirsutum* L.). *Spanish Journal of Agricultural Research*, 9(2): 460-472.
- Rad, M. N., M. A. Kadir, M. Rafii, H. Z. Jaafar, M. Naghavi and F. Ahmadi, 2013. Genotype environment interaction by ammi and gge biplot analysis in three consecutive generations of wheat (*Triticum aestivum*) under normal and drought stress conditions. *Australian Journal of Crop Science*, 7(7): 956-961.
- Yan, W., 2001. Ggebiplot—a windows application for graphical analysis of multienvironment trial data and other types of two-way data. *Agronomy journal*, 93(5): 1111-1118.