## Report on

3<sup>rd</sup> "Sino-Pak International Conferences on Innovations in Cotton Breeding and Biotechnology" September 04-06, 2019 Changji, China



# Organizer



Biotechnology Research Institute (BRI), CAAS, Beijing, China

# **Co-Organizers**



MNS University of Agriculture, Multan, Pakistan



Bahauddin Zakriya University, Multan, Pakistan

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#### SUMMARY

Cotton is a cash crop of Pakistan and China, having major role in the economy of both countries. China and Pakistan are 2nd and 5th largest producers of cotton in the world respectively. Cotton crop is facing abiotic and biotic stresses in the world which are affecting its yield and quality. To solve these issues a scientific collaboration was developed among the eminent scientists of MNS University of Agriculture (MNS UAM), Bahauddin Zakariya University (BZU), Cotton Research Institute, Multan, Pakistan with the Biotechnology Research Institute, Chinese Academy of Agricultural Sciences (BRI CAAS), Beijing, P. R. China.

To strengthen this collaborative relationship, 1<sup>st</sup> SINO-PAK International Conference on Innovations in Cotton Breeding and Biotechnology (ICICBB) was held at MNSUAM, Multan Pakistan, in November 2017 in collaboration with BRI, CAAS, China. The large group of Chinese scientists participated along with scientists of Malaysia, Turkey and Uzbekistan d to discuss the solutions for the issues of the cotton crop. After the successful conduct of 1st SINO-PAK, 2017, 2nd SINO-PAK ICICBB was also conducted on the same line to highlight the upcoming issues of cotton in the world, it was held at MNSUAM in collaboration with BRI, CAAS, BZU, CRI, PCPA and Government of Punjab on November 26-27, 2018. It was the second event in series after the successful conduct of 1st SINO-PAK ICICBB. The continuity of second event of ICICBB was the evidence of the success of this scientific collaboration between Pakistan and China. The aims of the conference were to bring together scientists, research scholars, ginners and progressive farmers on a single platform to share and enhance the knowledge about the solutions associated with the problems of the cotton crop. The conference was held to promote interdisciplinary dialogues regarding the contemporary issues in cotton.

It was agreed in the 2nd SINO-PAK, 2018 by the team leaders Prof. Dr. Zhang Rui from BRI CAAS and Prof. Dr. Asif Ali Vice Chancellor MNSUAM that the 3rd SINO-PAK International conference on Innovations in Cotton Breeding and Biotechnology will be organized by BRI CAAS, at China. The Biotechnology Research Institute, CAAS, China, organized 3rd SINO-PAK on Sep 04-06, 2019, at Changji, China. A delegate of scientists from MNSAUM, CRI, AARI, UAF and CEMB, Pakistan participated in the conference held at China.

#### **BACKGROUND AND INTRODUCTION**

Cotton is being grown, spun and woven into cloth for 3,000 years BC in Pakistan and China. Archaeologists found cotton fabric 5,000 years old at Mohenjo Daro, an ancient town in the Indus River Valley of West Pakistan. Today in Pakistan, it is a leading cash crop, producing 11.935 million bales during 2018. This crop has a 1% share in GDP and contributes 5.5 percent in agriculture value addition. It is being cultivated on an area of 2,699 thousand hectares. It is an annual crop grown in regions that experience climate variability. So, the high temperature, water scarcity and the salinization of the soil are major abiotic stress factors those limit cotton yield. The healthy cotton crop is unfortunately very attractive to insects throughout their whole growing period. Over 100 different types of pest's attack cotton which makes crop protection an important part of a cotton growers' job. In our region, the infestation of whitefly, pink bollworm and cotton leaf curl disease are the major damaging factors causing huge yield and quality losses. Farmers are extensively using pesticides to control these damaging tiny creatures. But the over use of pesticides is not a wise strategy, it is damaging environment and causing enormous harm to the farmer's friendly insects; the natural predators. Keeping in view the importance of cotton and the risks associated with its growth and development the MNSUAM realized to find solutions of these problems and helping growing community to ensure high production of good quality cotton, ultimately contributing to improve the national economy. For this purpose, linkages were developed with national and international, public and private institutes/organizations, especially the Biotechnology Research Institute, Chinese Academy of Agricultural Sciences, Beijing, P. R. China who has a major breakthrough in cotton biotechnology. It was decided to gather all cotton stakeholder with the aim to find a sustainable solution of problems faced by the cotton crop. It was the result of these linkages that 1st SINO-PAK International Conference on Innovations in Cotton Breeding and Biotechnology was held at MNSUAM. A team of Chinese scientists led by Dr. Rui Zhang from BRI, CAAS participated in the conference along with scientists from Uzbekistan, Malaysia and Turkey. A collaboration was developed between MNSUAM and BRI, China which opened new endeavors for researchers to share ideas and transfer of technology to enhance cotton productivity and profitability. The conference venue was Multan, a city that is the hub of agriculture with huge farmer's community, core cotton area of Punjab, Pakistan, a complex of cotton-related research institutes and private business companies and stakeholders. In the light of scientific talks and meetings, it was concluded that integrated pest management should be implemented to control the infestation of pink bollworm. Ultra-narrow row (UNR) spacing with nitrogen application at 150 kg/ha should be adapted to get maximum seed cotton yield and profit. The true potential of the genetic engineering should be exploited for the betterment of cotton crop. After one year of the first conference, a second SINO-PAK Conference on Innovations in Cotton Breeding and Biotechnology was held on November 26-27, 2018 at MNSUAM. It was successfully completed with the collaborative efforts of BRI, CAAS, MNSUAM, BZU, CCRI and Government of Punjab. It was two days conference where scientists from China, Pakistan, USA, Malaysia, and Iran talked on the following research themes i.e. Cotton Breeding, Cotton Genomics and Bioinformatics, Cotton Seed Technology and Sustainable Cotton Production. In the concluding session of the 2<sup>nd</sup> SINO-PAK, it was decided that the 3<sup>rd</sup> SINO-PAK will be held in China. So, the 3<sup>rd</sup> SINO-PAK was held at Changji, Xinjiang, China on September 04-06 which was organized by BRI, CAAS, China. The MNSUAM and BZU were co-organizers of the conference. The conference emphasized on collaborative efforts to solve common issues with cotton crop in both the countries. Eminent scientists from both the countries participated in the conference along with public and private sector stakeholders. The conference covers all aspects of cotton biology and breeding, generally attracting about 200 cotton scientists from Chinese and Pakistan to present and discuss their new discoveries. Changji is located in the northwest of Urumqi, the provincial capital of Xinjiang Uygur Autonomous region. Xinjiang is the largest cotton growing area at present in China. Changji is only 30 kilometers away from Urumqi International Airport, with direct flights to many countries, especially the countries along "one belt one road". Changji has beautiful scenery and has a long history, being the only route of the New North Road of Silk Road to the western region of Central Asia and Europe. Changji also has a strong cotton research community, especially in Western Research Center of Chinese Academy of Agricultural Science, a local host of 3rd Sino-Pak ICICBB. We will work together to make 3rd Sino-Pak ICICBB a memorable and productive conference.

## LIST OF MEMBERS OF DELEGATION FROM PAKISTAN

Sr.	Name of Participant	Designation	Affiliation
No			
1	Prof. Dr. Shafqat	Dean	MNS University of Agriculture,
	Saeed		Multan
2	Dr. Abid Mehmood	Director General	Ayub Agriculture Research Center,
		(Agric. Res.)	Faisalabad, Pakistan
3	Dr. Saghir Ahmed	Director	Cotton Research Institute (CRI,
			Multan)
4	Dr. Tayyab Hussain	Director	Center of Excellence in Molecular
			Biology (CEMB), Lahore
5	Dr. Masooma Naseer	Associate	Department of Plant Breeding,
	Cheema	Professor	University of Agriculture, Faisalabad
6	Dr. Zulqurnain Khan	Assistant Professor	Institute of Plant Breeding and
			Biotechnology, MNS University of
			Agriculture, Multan
7	Dr. Zia Ullah Zia	ARO	Cotton Research Institute (CRI,
			Multan)

## ARRIVAL AT CONFERENCE VENUE

The delegate was moved to the conference venue from Urumqi airport by bus. The delegate was arrived at conference venue, Yuanlin Hotel, Changji, after 30 minutes' drive. The delegate was warmly welcomed by the organizer of the conference, Prof. Dr. Zhang Rui.



Pakistani delegate welcomed by the host, Dr. Meng, BRI, CAAS

### FIRST MEETING WITH ORGANIZERS OF THE CONFERENCE

An introductory meeting was held on 04-09-2019 to discuss the current cotton situation and future prospects. It was decided to formulate a collaborative project under "One Road One Belt" for funding. All the organizers such as Prof. Zhang Rui, Dr. Liang, Dr. Meng, Dr. Way, Dr. Ali, Mr. Umar etc participated in the discussion. Dr. Abid Mahmood, Dr. Saghir Ahmed, Prof. Shaqat Saeed, Dr. Zia Ullah Zia and Dr. Zulqurnain Khan took the initiative to write the initial draft of the project. The minutes of the meeting are given below.

#### Minutes of the Meeting for Joint Research Projects

Collaboration with Biotechnology Research Institute CAAS China

September 4, 2019 Changji, China

With reference to our discussion about collaboration of Ayub Agriculture Research Institute (AARI) Faisalabad, MNS University of Agriculture Multan (MNSUAM) Pakistan with Biotechnology Research Institute (BRI) CAAS China following areas were identified for collaboration.

Prof. Zhang Rui pointed out that initial five-year phase of one belt one road is going to finalize in 2020 and now it is time to focus some project for short term and long-term collaboration. Prof. Dr. Shafqat Saeed pointed out that we have problem of Pink bollworm and whitefly in cotton. Prof. Zhang Rui offered that she has capacity and have specific genes/isolates of cotton for the management of pink bollworm. It will be good to develop a project and registered that gene in Pakistan as well as in China. There might be chances that it will be approved by Pakistani Govt. earlier, then we can use these constructs in Pakistani cotton varieties for management of pink bollworm. A summary of proposal was prepared at the same day and submitted to Prof. Zhang Rui for further discussion.

- 1. Germplasm exchange
- 2. Technology transfer
- 3. Mechanical picking
- 4. Pest and weed management
- 5. Training of scientists

## 5 Year Work Plan (2020-2025)

#### Germplasm exchange

Pakistan has strength in heat and CLCuD tolerant in cotton germplasm whereas Chinese scientists have very good quality of fiber germplasm. Therefore, germplasm will be exchanged between the two countries for improvement of cotton. The BRI has specific construct against Pink Bollworm management. The same will be tested in Pakistan and registered in Pakistan and China at the same time.

## **Technology transfer**

To start with, collaboration BRI under technology transfer will be made for pink bollworm resistance as it is one of the major issues of cotton in Pakistan. The BRI has developed construct for pink bollworm resistance. After successful lab/field trials in Pakistan the same will be registered in Pakistan. Later on, it will be expanded to advanced Bt and herbicide resistant technology.

## Pest management

IPM program will be launched with the collaboration of Chinese scientists for management of cotton pests particularly whitefly and pink bollworm.

## **Mechanical picking**

Chinese expertise as well as technology will be utilized for mechanical picking in Pakistan. Ultimately Pakistan will import Chinese picking machines as Pakistan feeling difficulty in manual cotton picking due to shortage of labour.

## **Training of scientists**

Total 4-6 Pakistani scientists will be trained in China for advanced biotechnological tools, pest management and cotton germplasm conservation.

Both the parties were agreed to submit projects under "One Road One Belt".



Pakistani delegate with Organizers of SINO-PAK ICICBB 2019

## **CONFERENCE PROGRAM** Wednesday, September 4, 2019

## Venue: Yuanlin Hotel

Time	Activity
9:30-21:00	Registration-Yuanlin Hotel Lobby
10:00-16:30	Sino-Pak ICICBB annual meeting
19:30-21:00	Dinner-TianChi Hall

## Thursday, September 5, 2019

#### Venue: Yuanlin Hotel

Time	Activity
9:30-14:00	Registration-Yuanlin Hotel Lobby
10:30-12:00	Opening Ceremony-TianChi Hall
12:00-12:30	Welcome Speeches
12:30-12:45	Photograph & Coffee Break

12:45-14:15	Keynote Speeches	Tianchi Hall	
Chair: Fuguang Li (Biotechnology Research Institute, CAAS)			
Shafqat Saeed (MNS-University of Agriculture, Multan)			
12:45-13:15	Xiaoya Chen (Shanghai Institute of Plant Pl	nysiology and Ecology, CAS)	
	Transcriptional regulation of cell wall looser	ing genes in cotton fiber	
13:15-13:45	Abid Mahmood (Ayub Agricultural Researc	ch Institute)	
	Breeding strategy for cotton improvement in	CLCV prone areas of Pakistan	
13:45-14:15	Rui Zhang (Biotechnology Research Institu	te, CAAS)	
	Cooperation makes a better tomorrow—from	n transgenic technology to	
	molecular breeding		
14:15-15:30	Lunch	Tianchi Hall	
15:30-17:30	Plenary Session I	Tianchi Hall	
Chairs: Xueyuan Li (Xinjiang Academy of Agricultural Sciences)			
Abid Mahmood (Ayub Agricultural Research Institute)			

 15:30-15:50 Xianlong Zhang (Huazhong Agricultural University) Developing biotechnological tools for functional genomics of cotton
15:50-16:10 Tianzhen Zhang (Zhejiang University) Mining of elite genes for high yield, superfiber qualities and heat tolerance and

their usages in improving Pakistan and China cotton cultivars

16:10-16:30	Shafqat Saeed (MNS-University of Agriculture, Multan)	
	Sustainable cotton production Sino-Pak collaboration opportunities	
16:30-16:50	Xiongming Du (Cotton Research Institute, CAAS)	
	A copy number variation of <i>GhDREB1</i> induced a causal mutation for extreme	
	dwarfism in upland cotton (G. hirsutum)	
16:50-17:10	Tayyab Husnain (Centre of Excellence in Molecular Biology, Lahore)	
	Exotic' genes expression in cotton for fiber quality improvement	
17:10-17:30	Yuehua Xiao (Southwest University)	
	Green fiber gene encodes a R2R3-MYB transcription factor promoting suberin	
	accumulation in the secondary cell wall of cotton fibers	
17:30-17:40 (	Coffee Break	
17:40-19:40	Plenary Session II Tianchi Hall	
	long Zhang (Huazhong Agricultural University)	
	<b>b Husnain</b> (Centre of Excellence in Molecular Biology, Lahore)	
17:40-18:00	Xinlian Shen (Jiangsu Academy of Agricultural Sciences)	
17.40-10.00	Introgression and exploitation of beneficial alleles from related diploid wild	
	species in <i>Gossypium hirsutum</i> background	
18:00-18:20	<b>Zhongxu Lin</b> (Huazhong Agricultural University)	
10.00 10.20	Genome-wide recombination landscape revealed the drive for genetic	
	improvement in cotton	
18:20-18:40	Jianbo Zhu (Shihezi University)	
10.20 10.40	Ectopic expression of the Pseudomonas aeruginosa KatA gene in cotton	
	improves its drought tolerance and yield under drought stress	
18:40-18:55	Xinhui Nie (Shihezi University)	
	Meta-QTL analysis of fiber quality traits in Upland cotton ( <i>Gossypium</i>	
	<i>hirsutum</i> ) based on SNPs	
18:55-19:10	Waqas Malik (Bahauddin Zakariya University)	
	Genome-wide characterization, evolutionary studies and expression patterns of	
	TALE transcription factor family in <i>Gossypium hirsutum</i> L.	
19:10-19:25	Qin Chen (Xinjiang Agricultural University)	
	Functional Genes exploration and Cyclophilin Family Genes Analysis During	
	Fibre Development of Sea Island Cotton	
19:25-19:40	Zia Ullah Zia (Cotton Research Institute)	

Evolution of MNH-872: a CLCuD resistant line developed through a threeway cross between (*G. arboreum*  $\times$  *G. anomalum*) and *G. hirsutum*.

## 19:40-21:00 Dinner

**Tianchi Hall** 

## Friday, September 6, 2019

10:00-12:00	Plenary Session III Tianchi Hall	
Chair	s: Tianzhen Zhang (Zhejiang University)	
Hongmei Cheng (Biotechnology Research Institute, CAAS)		
10:00-10:20	Huishan Guo (Institute of Microbiology, Chinese Academy of Sciences)	
	Cotton-Verticillium Interaction and trans-kingdom RNAi for cotton protection	
10:20-10:40	Yule Liu (Tsinghua University)	
	Molecular basis of cotton leaf curl Multan viral pathogenesis	
10:40-11:00	Guiliang Jian (Institute of Plant Protection, , CAAS)	
	Verticillium dahlia toxin to resistant and susceptible cultivar of Gossypium	
	hirsutum leaf Proteomic by iTRAQ	
11:00-11:20	Saghir Ahmad (Cotton Research Institute, Multan)	
	Introgression of Cotton Leaf Curl Virus (CLCuD) resistant genes from G.	
	arboreum to G. hirsutum.	
11:20-11:40	Zulqurnain Khan (MNS-University of Agriculture, Multan)	
	Comparative study of TALEs/TALENs and Cas9/dCas9 for virus resistance in	
	cotton	
11:40-12:00	Masooma Nasir Cheema (University of Agriculture, Faisalabad)	
	Consequences of poor stewardship of Bt cotton in Pakistan	
12:00-12:20	Kamal Ghasemi Bezdi (Agricultural Research, Education and Extension	
	Organization, Cotton Research Institute of Iran)	
	Not confirmed Yet	
12:20-12:30	Coffee Break	
12:30-13:45	Plenary Session IV Tianchi Hall	
Chair	s: Xiongming Du (Institute of Cotton Research, CAAS)	

Saghir Ahmad (Cotton Research Institute, Multan)

12:30-12:50 Xueyuan Li (Xinjiang Academy of Agricultural Sciences) The Future of Xinjiang Cotton Industry under the New Era their usages in improving Pakistan and China cotton cultivars

12:50-13:10	Yabing Li (Institute of Cotton Research, CAAS)		
	Eco_physiological mechanism of mechanized cotton production of China		

- 13:10-13:25Xiaoguang Shang (Nanjing Agricultural University)Comparative analyses reveal the genetic basis of fiber improvement in<br/>cultivated versus semi-domesticated cotton (Gossypium hirsutum L.)
- **13:25-13:35** Iftikhar Ali (Institute of Cotton Research, CAAS) Fine-mapping of *qFS21* and candidate gene identification
- 13:35-13:45Abdul Rehman (Cotton Research Institute, CAAS)Identification and characterization of *HSP70* gene family in cotton

13:00-14:15	Closing Ceremony Tiar	nchi Hall	
Chair: Rui Zhang (Biotechnology Research Institute, CAAS)			
Shafqat Saeed (MNS-University of Agriculture, Multan)			
13:00-13:05	Play National Anthem of People's Republic of China		
	Play National Anthem of Islamic Republic of Pakistan		
13:05-13:15	Prof. Dr. Tayyab Hussain		
	Director of Center of Excellence in Molecular Biology, I	Lahore	
13:15-13:25	Dr. Zulqurnain Khan		
	MNS-University of Agriculture, Multan		
13:25-13:35	Prof. Dr. Zhang Rui		
	BRI, Chinese Academy of Agricultural Sciences, Beijing	, China	
14:15-15:30	Lunch	Tianchi Hall	
15:30-19:30	Cotton Farm and Seed Factory Visiting		
19:30-21:00	Dinner	Tianchi Hall	
Saturday, Sej	ptember 7, 2019		

Whole Day Departure

#### **INAUGURAL CEREMONY**

Inauguration of ICICBB was started at 10:00 am at Yuanlin Hotel hotel. More than 100 Chinese scientists participated throughout the China and following leaders addressed the participants. The opening ceremony of the conference was chaired by **Prof. Guoqing Sun**, Director of Research Management Office, BRI, CAAS. The professor welcomed all the participants of the ICICBB in the opening ceremony of the conference. **Prof. Hao Weiping**, Deputy Director of Department of International Cooperation of CAAS extended the agenda of the conference. He also welcomed the participants with the hope that the conference will be very beneficial for all the participants.

**Mrs. Ma Lijun**, Deputy Director of the Changji Hui Autonomous Prefecture People's Congress Standing Committee welcomed the participants from local government of Changji. She said these type events are appreciated by the local government which would be beneficial for the country and local community. **Prof. Li Xinhai**, Director General of the BRI, CAAS emphasized on the theme of the conference as host of the organizing institute. He further said that his institute is enthusiastic to organize this type of international events. Academician, **Prof. Chen Xiaoya**, Institute of Plant Physiology and Ecology, Chinese Academy of Sciences also talked in the opening ceremony to congratulate the organizers and participants of the participants to the conference. As the Chairman of the organizing committee, he extended his feelings of gratitude to the organizing committee for organizing the conference. He was thankful to the collaborators of the conference; MNSUAM and BZU, Pakistan. He was especially grateful to the Pakistani delegate for participating in the conference.



Dr. Abid Mehmood, Director General Research, Punjab, Pakistan, his remarks, in appreciated the efforts of MNSUAM, CAAS, Beijing China, BZU, Multan and CCRI, Multan for providing an opportunity to the researchers, academia, growers, public and private sector seed producers, students and other stakeholders to sit together and discuss the issues faced by cotton crops and finding



solutions of the problems. He hoped that the effects of these efforts will leach to the end users. If such conferences will be conducted on yearly basis there is no doubd that we will overcome all issues related to this King Crop. He further presented the cumulative recommendations derived from all the talks delivered during five technical sessions. These recommendations are given above.

#### PRESENTATIONS OF THE PARTICIPANTS FROM PAKISTAN

**Dr. Abid Mehmood**, Director General, Agriculture Research, Punjab, presented a comprehensive outlook of the cotton crop in Pakistan. Cotton is a crop of extreme importance in Pakistan and provides livelihood to millions of people directly or indirectly. Cotton alone contributes 51% of the total foreign exchange, 5.1% in value addition and



1% in GDP of the country. Pakistan ranks second in yarn export, third in consumption, yarn production, cloth production & cloth export and fourth in cotton production among the 60 cotton producing countries of the world. Cotton is primarily a fiber crop yet it also fulfils about 70% of the total requirement of edible oil of the country. Cotton is grown on an area of about 3 million ha with about production of 12 million bales every year. Punjab province contributes about 70% of the total production of the country. Cotton Research Institute is one of main and oldest research Centre in Pakistan which was established in 1907 in Faisalabad and the directorate was shifted to Multan in 2017. It has developed 59 cotton varieties with an economic contribution of Rs. 1400 billion to the national economy since 1980. First variety of upland cotton was 4-F which was introduced in 1914 developed at CRS Faisalabad. Seed cotton Yield potential of 4-F was 890 kg/ha with Got 32%, staple length 20.8mm, fiber fineness 5.8µg/inch and 310 days were required till the physical maturity of this variety. After that extensive research in cotton breeding resulted in major genetic improvement. Among non Bt varieties the most popular cultivars were MNH-993, NIAB-78, S-12 and MNH-786 with significant improvement in yield and fiber traits. Commercialization of Bt varieties carrying Cry1Ac gene and MON-531 event started in 2010 and uptill 20 Bt varieties have been developed by CRI Multan and its allied research stations. Most popular Bt varieties among the cotton growing community are MNH-886, FH-142, NS-121 and IUB-13. Now the varieties i.e. FH-Lalazar are early maturing which take 196 days for maturity with a genetic gain of 37%. Yield potential has increased to 6000kg/ha with a genetic gain of 574% over 4-F. Bt varieties are now being cultivated on around 95% area in Pakistan and 99% area in Punjab.

**Dr. Saghir Ahmad** Director Cotton Research Institute Multan gave a talk on "Introgression of Cotton Leaf Curl Virus (CLCuD) resistant genes from *G. arboreum* to *G. hirsutum*". He said that Cotton leaf curl virus disease (CLCuD) is one of the major factors putting primary limit on cotton productivity particularly in Punjab



province of Pakistan with an annual loss of 2-3 million bales amounting to US\$ 612-918 million to the economy of the country. The same disease has also been reported in China in 2008 in Nanning city, Guangxi, and Uzbekistan in 2014. Epidemic form of this disease in Pakistan occurred during 1992-1996. Research work on CLCuD started in 1993 and germplasm was imported from abroad and two lines LRA-5166 and CP-15/2 (Indian origin) were found resistant to Khokhran virus-dominant at that time. First CLCuD resistant variety CIM-1100 was evolved in 1996. Later on, a number of CLCuD resistant varieties were developed by different institutes in Punjab during 1996-2000. In the wake of resurgence of this disease during 2002 in Burewala Vehari the resistance was broken down into all varieties including whole germplasm became susceptible to this disease. Up till now no CLCuD resistant variety of G. hirsutum has been evolved by any institute in Pakistan. However, G. arboreum having A-genome has shown resistance against CLCuD. Therefore, we planned to introgress CLCuD resistant genes from G. arboreum into G. hirsutum through interspecific hybridization in 2009. Tetraploid of G. arboreum were developed through Cholchicine application to facilitate hybridization and production of fertile progenies. A few CLCuD resistant lines through hybridization between G. arboreum and G hirsutum followed by successive backcrossing have been developed by the team at Cotton Research Institute Multan. MNH-1050 is the first promising line/product obtained through introgression of the two species G. arboreum and G. hirsutum. This line possesses harmonious combination of seed cotton yield and fiber traits, i.e., seed cotton yield = 4000kg/ha, Ginning out-turn = 42%, Staple length = 29.7 mm, Mike = 4.6  $\mu$ g/inch, Fiber Strength = 32.0 g/tex against the commercial cultivar IUB-13 possessing lint yield = 2356kg/ha, Ginning out-turn = 41.95%, Staple length = 28.8 mm, Mike =  $4.7 \mu \text{g/inch}$ , Fiber Strength = 28.0 g/tex. This line will be included in National trials in 2019 for its approval as a variety.

**Prof. Dr. Shafqat Saeed** greatly appreciated the Chinese scientists for their collaboration and helping the world particularly Pakistan in combating challenges encountered by the Cotton crop. He underlined the need to find sustainable solutions for problems incurred by changing climate scenario. The



development of resistance against heat, sucking insects pests especially whitefly, pink bollworm and begomoviruses induced Cotton leaf curl disease should be achieved by coordinated research activities beyond borders. Professor Dr. Asif Ali shared the success story of the management of Pink bollworm by the MNSUAM. The Pink bollworm is a devastating pest of Cotton drastically reducing yield and quality. He told the audience about the management strategy (the use of PB ropes) involving only one spray in whole cropping cycle enormously reducing the number of sprays for Pink bollworm and whitefly which were normally 12 to 16 per cropping cycle. He further said that Bt technology is very important for Pakistan. The country witnessed major decline of cotton production in 2015 due to pink bollworm outbreak. This was the result of all above mentioned reasons, which contributed towards the development of resistance in the target pest. Farmers are now adapting double gene cotton, which need to implement all precautionary measures for the prolonged and durable performance of this technology against target pests. He further emphasized in Chinese collaboration for technology and germplam tranfer. The collaborative projects are needed to cope with the problems in cotton.

**Dr. Tayyab Hussain** presented his work entitled "'Exotic' genes expression in Cotton for fiber quality improvement". Cotton fiber is a differentiated single cell trichome that originates as an extension of ovule epidermal cell. Improvement in fiber quality of cotton is mainly controlled by the genetic traits it holds and their expression. The aim of this study was to transform different fiber related genes



through Agrobacterium mediated transformation into local cotton varieties for improvement of fiber quality. Expression of bacterial cellulose genes (acsA and acsB) in a local cotton variety under fiber specific promoter (GhSCFP) resulted in increased in fiber length (upto 17. 52%), increased fiber strength (upto 26.45%) and decreased fiber micronaire value (upto 22.41%) in the transgenic lines as compared to the control non-transgenic cotton plant lines.Agrobacterium mediated transformation of Flavonoid 3'5'hydoxylase (F3' 5' H) and Dihydro flavonol 4- reductase (DFR) in to cotton increased staple length (upto20.1%), fiber strength (upto32.7%) and micronaire value (upto 4). Similarly, HOX3 and WLIM5 transformed under fiber specific promoter resulted in increase of staple length (upto 26%), strength (8.7%) and micronaire value was found to be improved (upto5.2) in G. arboreum. Transformation of Sucrose synthase (SuS) gene under constitutive promoter also showed an increase in fiber length (up to 11.7%), fiber strength (18.65%), cellulose contents (28%) whereas value of micronaire recorded was 3.5. SEM analysis of mature fiber revealed improvement in fiber surface smoothness as compared to control non-transgenic cotton plants. The material developed can best be utilized in breeding programme for possible gene pyramiding to have combinational impact on fiber quality.

**Dr. Zia Ullah Zia** is a breeder at CRI, Multan. He presented his work on "Evolution of MNH-872: a CLCuD resistant line developed through a three way cross between (*G. arboreum*  $\times$  *G. anomalum*) and *G. hirsutum*". He talked about his previous work of breeding in cotton to develop CLCuD resistant cotton variety. Domesticated cotton species and its wild relatives belonging to genus Gossypium



constitute more than 50 members in this genus. These members are genetically very diverse ranging from tree sized plants to small shrubs throughout the world. G. anomalum (2n = BB = 26) is a diploid and wild member of this genus from B-genome, while G arboreum (2n = AA = 26) is a diploid and cultivated specie from A-genome and both are resistant to CLCuD. A cross between G. arboreum × G. anomalum was attempted followed by colchicine application for doubling of chromosome. This amphiploid (2n = AABB = 52) was crossed with G. hirsutum (2n = AADD = 52) at Cotton Research Institute Multan. Backcrossing using G. hirsutum as recurrent parent was followed till BC4F1 for integration of CLCuD tolerant genes in G. hirsutum. Selection cycle was started from BC4F2 for desirable traits and continued till BC4F6. MNH-872 was selected in BC4F7 for displaying a harmonious combination of yield and fiber traits along with resistance against CLCuD. Fiber traits of MNH-872 are; GOT = 42.1%, Staple length = 32.0 mm, Mike = 4.5 µg/inch and Fiber Strength = 39.4 g/tex. This advance line will be included in yield trials in 2019 for varietal approval procedure.

**Dr. Masooma Naseer Cheema**, Associate professor, PBG, UAF, presented her work entitled "Consequences of poor stewardship of Bt cotton in Pakistan". Bt cotton technology has revolutionized the production in the developed countries, but its adoption in the developing countries didn't perform up to



the mark. Comparing the situation in both scenarios showed non-stewardship of this technology in the developing countries, where the system was not supervised and regularized as it was in the developed world; ranging from seed marketing to the policy matters. The stealthy diffusion of single gene Bt cotton in Pakistan, during the first decade of this century, is the first indicator of poor monitoring over the spread of this technology. After the Bt cotton varieties were formally approved in 2010, another round of fooling the farmers started with the selling of unapproved lines and non-certified seed in the seed market. Non-availability of the system monitoring resistance development in target pests further worsened the situation. Non placement of the system for testing meeting the legalities of the transgenic plant's seed also contributed towards the factors of cotton decline in the country over couple of years. Another major cause is the nature of single gene Bt cotton, which has spatio-temporal expression variability and is highly affected by the environment. The country witnessed major decline of cotton production in 2015 due to pink bollworm outbreak. This was the result of all above mentioned reasons, which contributed towards the development of resistance in the target pest. Farmers are now adapting double gene cotton, which need to implement all precautionary measures for the prolonged and durable performance of this technology against target pests. But the good news is about the performance of Cry2Ab gene. It didn't show variable expression under our local climatic conditions, instead it has consistent and higher expression level as compared to Cry1Ac gene.

**Dr. Zulqurnain Khan,** Assistant Professor, Institute of Plant Breeding and Biotechnology, MNSUAM talked on "Comparative study of TALEs/TALENs and Cas9/dCas9 for virus resistance in cotton". Dr. Khan said that cotton leaf curl disease (CLCuD) is one of the major factors for low yield of cotton in



Pakistan. Engineering resistance in cotton cultivars against CLCuD through RNA-directed Cas9 is a powerful tool to tackle the viral disease in cotton. Previously, we used TALEs, TALENs and dCas9 to target cotton leaf curl virus and got promising results. We have targeted coding and non-coding regions of viral DNA individually with Cas9/TALENs and dCas9/TALEs and found promising potential target sites for viral interference. On basis of screening results of gRNAs tested in transient assay in *Nicotiana benthamiana*, we designed multiplex gRNA for targeting three most promising sites simultaneously with multiplexed Cas9. We found that targeting non-coding regions of virus is more effective for virus suppression than targeting coding regions. In transient assay, we found 70-90% decrease in accumulation of virus. Cotton transformation with multiplex gRNAs is under progress. DsRED was used as a visible marker in stable transformation of cotton to optimized the transformation protocol.

## PROJECT DISCUSSIONS AND RESEARCH COLLABORATIONS

- Project was written with Zhang Rui's group (BRI, CAAS, China)
- Discussion on future collaborations was made successfully with Tianzhen Zhang group (Zhejiang University, Wuhan, China).
- MOU and MTA Agreement was signed with Prof. Zhongxu Lin (Huanzhong Agriculture University, China)
- Cotton germ plasm was taken from Prof. Zhongxu Lin for further studies in Pakistan
- Meeting was conducted with Prof. Xin-Hui Nie, Director/Secretary of the department of agriculture, Shihezi University for research collaborations and cooperation.
- More emphasis was given to "One Road One Belt" program for writing projects to hunt grants.

#### Minutes of the meeting with Tianzhen Zhang, Zhejiang University, Wuhan, China

A very successful meeting with conducted with Prof. Tianzhen Zhang. Prof. Zhang is an eminent scientist working on cotton breeding and biotechnology at Zhejiang University, Wuhan, China. All members of the Pakistani delegate were present in the meeting. During the discussion in the meeting, emphasize were given on the following points;

1. Genome assembly will be done taking a reference genome from Pakistan (GWAS, GBS)

2. Sequencing of cotton germ plasm will be done especially Pakistani genotypes.

3. Heat tolerance lines will be shared from Chinese partner for their acclimatization in Pakistan.

4. Triple gene (Cry1Ab, Cp4, 3x) construct and transgenic cotton will be shared from Chinese partner.

5. It was decided that joint labs will be established at MNSUAM, CRI and Zhejiang for further research on cotton breeding and biotechnology.

6. Chinese varieties will be provided for backcrossing.

7. Joint proposal will be written for submission to CPEC.

8. Tripartite Agreement Draft will be prepared by the Pakistani partner after the arrival.

9. Training programs for graduate students and faculty will be launched under exchange program to strengthen the research and for capacity building of the researchers.



Meeting of Pakistani delegate with Prof. Tianzhen Zhang, Zhejiang University

## VISITS

### Visit of Xinjiang Agriculture Expo, Changji, Xingjiang

The visit was arranged by the organizers of the conference. The delegate was welcomed by the official of the Expo Center. A visit of the green houses and fields was hosted by the officers of the Expo Center. The Center have many demo plots of the crops such as cotton, corn etc and vegetables and ornamental plants. The green houses have well maintained plants which were grown by different private and public sector organizations. The center has stateof-the-art facilities for acclimatization of the exotic plant species. The cut flowers of different types were well grown and were demonstrated for commercialization. The center also has big shopping store to market its own products for public.



Visit of the Xinjiang Agriculture Expo, Changji, Xinjiang with conference organizers.

#### Farmer's Field Visit

A farmer field visit was arranged by the organizers for all the participants at the end of the conference. Cotton fields grown by different farmers at the out skirts of the Changji city were very well cultivated. It was told by the farmer that the land is taken from the government on lease. The amount of the lease is up to 600 RMBs/acre. The cost of production for one acre is 1600 RMBs while 400 RMBs subsidy is provided by the government. It was told by the guide that the average production of local cotton variety was nearly 50 Monds. Net profit of the crop is nearly 3000-4000 RMBs. It was observed that Chinese cotton have no significant damage of insects and diseases. The temperature for the crop was ideal in Changji. All the cotton fields were irrigated through drip irrigation system. Chinese scientists told that Xinjiang province produce 70% of the total cotton of China.



Farmer's field visit at Xinjiang

#### Visit of Join Hope Seed Company

After farmer field visit, the participants moved to the seed company "Join Hope" which was on the other side of the city. The participants were warmly welcomed by the CEO of the company. It was told by the director of the company that the company was started in 2007 with investment of 700-800 million RMBs. To date, the company has assists of 1.8 billion RMBs. Major seed developed by the company is of Wheat, Maize and Cotton. It was told that the annual seed supply is about 8000 tons. The participants visited seed processing units of the company. It was told that the automated machinery for seed processing, grading and packing was imported from Germany. The director of the company also visited cotton and corn field of the company. Seeds of all the varieties was displayed in the expo of the company which was visited later. At the end of the visit, a meeting was arranged by the officials of the company with the participants of the conference. DG Res Dr. Abid Mahmood asked the company to come to Pakistan to explore their business and to provide their high productive seed to Pakistani farmers. It was decided that public private sector cooperation will be encouraged to solve the issues of low production and low seed quality in both the countries. It was told be the director research of the company that the company is working with 30 government sector institutes and universities within China. Moreover, the company also doing business in more than twelve countries of the world and supplying seeds of cotton, wheat and maize.



## **CLOSING CEREMONY OF THE CONFERENCE**

The closing ceremony was chaired by Prof. Zhang Rui and Prof. Shafqat Saeed. Dr. Tayyab Hussain gave the closing remarks. He appreciated the efforts of the organizing committee of the conference. Dr. Hussain congratulate Prof. Zhang Rui and her team on successful



conduct of the conference. He said that this SINO-PAK platform is very useful and beneficial for the farmers, researchers, private stakeholder, academician and students to understand the issues in cotton and to know the latest research being done for cotton improvement. Prof. Shafqat Saeed also congratulate the team on organizing such a tremendous event. He also said that these events will increase the bilateral cooperation in other fields as well. Prof. Zhang Rui said that SINO-PAK cooperation is need of the hour to solve common problems for progress and development in both the countries; China and Pakistan. She said that she ambitious for future collaboration with MNSUAM to do joint projects.



Presenting memories of 2<sup>nd</sup> SINO-PAK, 2018 to Prof. Dr. Zhang Rui

#### Success Story of 1<sup>st</sup> and 2<sup>nd</sup> SINO-PAK Conferences

The Success Story of 1<sup>st</sup> and 2<sup>nd</sup> SINO-PAK Conferences was presented in the closing session of the conference by Dr. Zulqurnain Khan. He told about the history of the program. He said that the program was initiated in 2017 with the meeting and collaboration of the two leaders Prof. Asif Ali, Vice Chancellor, MNS University of Agriculture, Multan, Pakistan and Prof. Zhang Rui, BRI, CAAS, Beijing, China. Both the parties decided to hold a conference emphasizing cotton breeding and biotechnology to provide importance



to issues in cotton and to make bilateral efforts for solution of the problems. Increasing cotton production with good quality and sustainability was the moto of the program. Talking about the previous outcomes of the conferences held in 2017 and 2018, Dr. Khan told the audience that joint lab and join projects have been started under SINO-PAK collaboration. A lab at MNS University of Agriculture, Multan, Pakistan, was established which is now in working conditions. Moreover, many visits have been made across by scientists of both countries. Collaborative projects have been submitted for to enhance cotton yield and quality. At the

end, Dr. Khan appreciated and prayed for future achievements and successful conduct of the conferences. Moreover, he also told about organizing the 4<sup>th</sup> SINO-Pak Conference at Pakistan as conference secretary. The organizers of the 3<sup>rd</sup> SINO-PAK, 2019 were highly appreciated and congratulated on successful conduct of the conference. Chinese and Pakistani participants showed their interest and support to continue the SINO-PAK conference program with bilateral hard



work and cooperation. It was decided that the scientists of other countries will also be invited to participate in the conference to make the platform open for world scientists. Public and private cooperation is needed to organize such events. Moreover, suggestions were given to highlight the program on print, electronic and social media to gain more attention and audience.

#### **EXECUTIVE SUMMARY**

The third Sino-Pak International Conference on Innovations in Cotton Breeding & Biotechnology just concluded in China's Xinjiang Uygur Autonomous Region. The conference attracted over 100 experts and scholars on cotton field. They made communication and discussions on related aspects, such as, cotton germplasm, transgenic cotton against insect and herbicide, transformation technology as well as the mechanized planting and picking of cotton.

The experts of both countries agreed the China-Pak Joint Laboratory for Cotton has made plentiful achievements after three years' construction. They hope to further deepen cooperation field and will strive to promote the development of the two countries' cotton industries. The success holding of the third Sino-Pak International Conference on Innovations in Cotton Breeding & Biotechnology not only pushes ahead the sci-tech communication and cooperation in agriculture between the two nations, but also injects new vitality for the Belt and Road Initiative.

The third Sino-Pak International Conference was jointly sponsored by the Biotechnology Research Institute of Chinese Academy of Agricultural Sciences, MNS-University of Agriculture and Bahauddin Zakariya University. The China-Pak Joint Laboratory for Cotton is under the China-Pakistan Economic Corridor (CPEC). Now the first stage of CPEC has almost been completed. For the second phase of CPEC, the agriculture will be one of the focuses among others.

#### Departure from Changji (Urumqi) and Arrival in Pakistan

The participants were taken to the Urumqi airport from Changi by bus to take the flight CZ6017 of China Southern on 07 Sep, 2019 at 8:15 am. The participants arrived safely at Lahore airport around 10:00 am according to local time.



Group photo of the participants of the 3<sup>rd</sup> SINO-PAK ICICBB, 2019