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Cotton Germplasm Resources in China

Yinghua Jia, Junling Sun and Xiongming Du

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1. Introduction

1.1. History of cotton germplasm research in China

China is one of the oldest countries that was found to have a long history of growing cotton. The remotest record was searched in one of the Chinese historic literature book *Shang Shu.Yu Gong* (about 300 BC), which described that cotton named *Jibei* was used to weave colorful cloth by the native people in the island of South China [1-2,4]. The primary species of cotton were two diploids including *G. arboreum* L. in South China and *G. herbaceum* L. in West China, but *G. arboreum* L. was only kept growing in the south of the Five Ridges area, Hainan Island and Yunan until Huang Daopo (about 1245–1330AD) improved the technology of waving cotton fiber and brought it to Songjiang in Shanghai from Yazhou in Hainan. Then, *G. arboreum* L. was widely cultivated in the mid and lower reaches of the Yangtze River and was spread to North China in Ming Dynasty. *G. herbaceum* L. was firstly found to be planted in Xinjiang, but it was not popularized across the country, and was only cultivated in Xinjiang and Gansu Provinces in the past years. *G. hirsutum* L. was firstly imported to China by Zhang Zhidong in 1892-1893 and was cultivated in Hubei. *G. barbadense* L. was imported to China earlier than *G. hirsutum* L. and was mainly in Yunan province as perennial cultivars [3-5]. Now China is the largest producer and consumer of cotton in the world. According to the World Bank, from 2005-2007, China produced 28.4 to 37 million bales of cotton and consumed 45 to 51.5 million bales in the country's domestic textile mill industry. On average, cotton was produced on ~5.6 million ha in China within three predominant growing areas including Xinjiang area, the Yellow River area, and the Yangtze River area. Similar to India, China's adoption of Bt-hybrids and cultivars have resulted in marked yield increases in recent years [6-7].

The Chinese cotton germplasm collection located at the Institute of Cotton Research of Chinese Academy of Agricultural Sciences in Anyang. A medium-term working collection located at Anyang, and a long-term collection located at Beijing, which was established in 1958. An *in*

in vivo collection of wild species was housed at Hainan Island. The earliest medium-term germplasm bank for cotton was built in Baibi town in 1959-1960, and the seeds must be reproduce every three years, which could preserve the cotton seeds for 4-5 years because of the primitive condition. Then a semi-automatic temperature controlling medium-term germplasm bank was rebuilt in 1979-1982, which was enlarged for the storage area and was constituted of two rooms. This semi-automatic germplasm bank could kept the seeds for more than ten years at 0°C. An automatic temperature controlling medium-term grmplasm bank was built in Anyang city in 2000-2002, which could prolong the preservation of the seeds for fifteen years at 0+2°C below 50% relative humidity. Now a new germplasm bank has been rebuilt based on the old building in Baibi town in 2011-2012, which is larger and can hold more than 30000 cotton germplasm lines at the temperature 0C and 50% relative humidity. The *in vivo* collection garden was built in Yachen, Shanya, Hannan in 1979 due to the warm and moist weather in the winter. Till now, more than 36 wild species were nursed in that place.

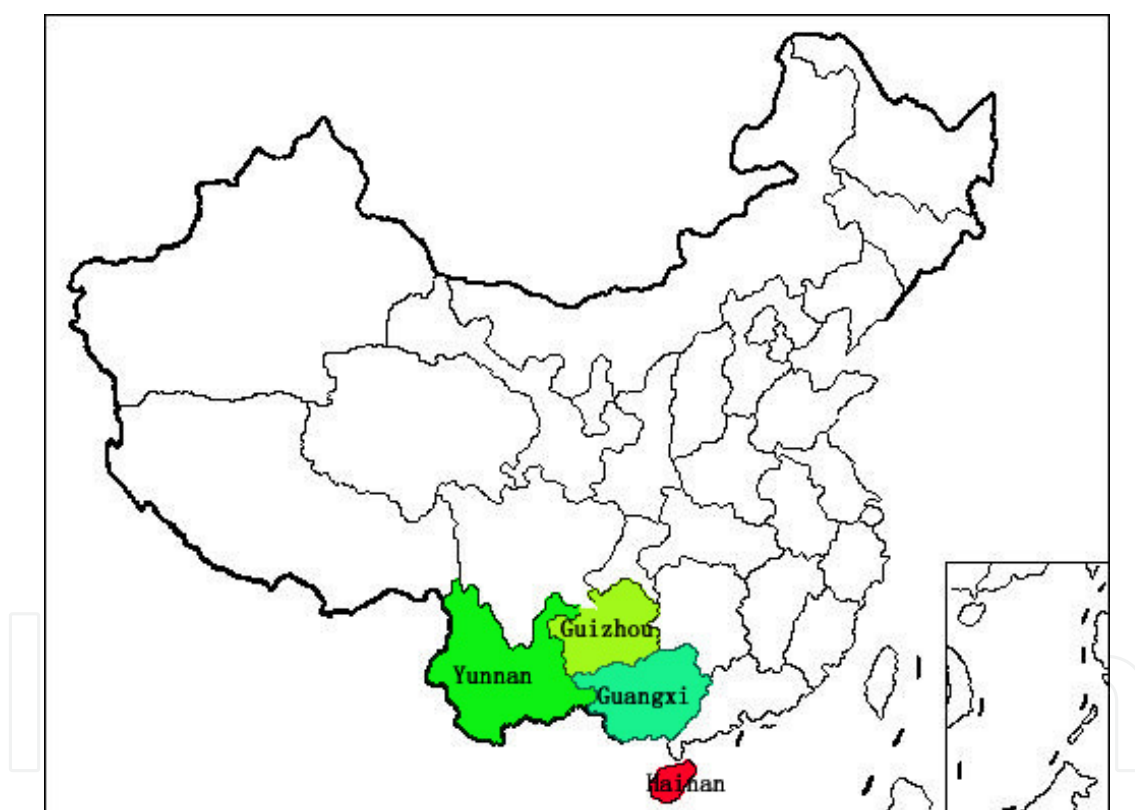


Figure 1. Map of exploration of *G. arboreum* L. in south-west china

Six professors have been in charge of the germplasm bank and *in vivo* garden from 1958 to 2013, and made important contributions to the development of cotton germplasm in China. Prof. Meng Shiheng was the first researcher in charge of cotton germplasm work during 1957-1960, and he established the early system of preservation of cotton, including information collection of pedigree and specimens. Xiang Xianlin continued the work and built seven ecological conservation sites from 1958-1989. Xiang edited and published the first two

catalogues of cotton germplasm in China [8-9]. She organized the collection, conducted evaluation and exploration of *G. arboreum* L. in the south-west China. Later Prof. Liu Guoqiang was in charge and kept on the work of cotton germplasm. He arranged to classify and evaluate all the cultivars in the germplasm bank from 1981 to 2001. Prof. Liu edited and published the latest catalogue books of cotton germplasm in China [10-12]. Prof. Du Xiongming added molecular method in the evaluation of the diversity of cotton germplasm, and identification the genes related to elite agronomic traits in recent years [13]. Prof. Wang Kunbo was in charge of the preservation of wild species of cotton. He built the nursery station for *in vivo* preservation, and evaluated the characters of species with FISH and molecular method [14].

2. Collection of cotton germplasm in China, exploration, exchange with other countries

From last century, collection in homeland has gone through three stages of development. The first stage was the years before the 1949. Wang Shanquan and Feng Zhefang firstly started to collect the *G. arboreum* L., from thirteen provinces in the country including Jiangsu, Zhejiang, Liaoning, Shangdong (in 1922-1923) etc., and acquired 112 lines with diversity phenotypes [1]. Later, Central Agricultural Research Institute continued the work and collected 1200 *G. arboreum* L. But most of these lines were lost during World War Two. The second stage was the middle of the last century. In 1956-1957, the Ministry of Agriculture announced to support for collection of the landraces across the country assisted by the local government. Until 1961, almost 1007 cotton lines were received from all over the country, including 697 *G. hirsutum* L., 25 *G. barbadense* L., 256 *G. arboreum* L., one *G. herbaceum* L. and five wild cottons. Regular collection expeditions were also organized at this stage. The crop explorations have covered most of South-west China. In 1975, Xiang Xianlin, Ma Xijing and Wang Biying explored Luxi, Yinjiang, Longchuan, Jinghong, Mengla in South-west of Yunnan Province. In 1980, Xiang Xianlin explored Yaxian, Linshui, Baoting, Yuedong, Baisa, Danxian, Dongfang, Wenchang in Hainan province with the help of local government. In 1983, Xiang Xianlin, Zhang Zhujie, Yang Shiquan explored the south of Guizhou Province, and in the same time Liu Guoqiang and Shen Duanzhuang explored the north of Guangxi Province. In this stage, the agricultural institutes in the main provinces also kept their cultivars genetic stocks and sent to the long-term germplasm bank in Beijing after evaluation till 1995. The third stage was years after 2000. In 2002-2013, lines preserved in local agricultural institutes of Xianyang, Tangshan, Xiaoshan, Jiuquan, Bazhou, Binchuan, were sent to Institute of Cotton Research of Chinese Academy of Agricultural Sciences for long-term preservation. Explorations of landrace cotton were also carried out in this stage. Profs. Wang Kunbo and Du Xiongming organized the explorations in Yunan, Guizhou, Guangxi Province separately from 2002 to 2013 (Table 1).

Years	Locations	Acquirement	Explorer and institute
1922-1933	Thirteen provinces in china, including Shangdong, Jiangsu, Zhejiang, Liaoning	112 <i>G. arboreum</i> L. were collected	Wang Shanquan, Feng Zhefang
1939-1949	All over the country, including Sichuan, Yunanan	1200 <i>G. arboreum</i> L. were collected	Wang Shanquan, Feng Zhefang, Xi Yuanling, Chenren et al in Central Agricultural Institute
1952		73 <i>G. arboreum</i> L. were collected	Huabei Agriculture Research Institute
1956-1961	All over the country		Agriculture Ministry and Xiang Xianlin
1980	Hainan Province, including Yaxian, Baoting, Linshui, Yuedong, Baisha, Danxian, Dongfang, Wenchang	78 accessions including <i>G. arboreum</i> L., and <i>G. hirsutum</i> L.	Xiang Xianlin in Institute of Cotton Research of Chinese Academy of Agricultural Sciences, science and technology department in Hiannan province.
1983	Guizhou province, including Kaili, Chongjiang, Rongjiang, Sandou, Luodian, Wangmo, Guanling, Zhenning	36 <i>G. arboreum</i> L. and 16 <i>G. hirsutum</i> L.	Xiang Xianlin, Zhang Zhuxian in Institute of Cotton Research of Chinese Academy of Agricultural Sciences, and Yang Shiquan in Guizhou
1983	Guangxi province, including Donglan, Bama, Nandan, Sanjiang, Fuchuan, Guiping, Debao, Jingxi, Pingguo	18 <i>G. arboreum</i> L. and 22 <i>G. hirsutum</i> L.	Liu Guoqiang, Fu Huaqin in Institute of Cotton Research of Chinese Academy of Agricultural Sciences, Sheng Duanzhuang in Jiangsu Academy of Agricultural Sciences
2002	Guangxi, Guizhou, Yunan province	89 <i>G. arboreum</i> L., 43 <i>G. hirsutum</i> L. and 18 <i>G. barbadense</i> L.	Wang Kunbo, Liu Fang et al in Institute of Cotton Research of Chinese Academy of Agricultural Sciences
2005	Yunnan, Guangxi province, including Mojiang, Jiangcheng, Kaiyuan, Maguan, Funing, Napo, Leye, Longlin, Tiandong, Pingguo, Daxin, Debao, Shangsi	Six <i>G. arboreum</i> L., two <i>G. barbadense</i> L.	Liu Guoqiang, Jia Yinhua in Institute of Cotton Research of Chinese Academy of Agricultural Sciences
2009	Guizhou, Guangxi province	16 <i>G. arboreum</i> L.	Du Xiongming, Zhou Zhongli, Jia Yinhua, Sun Junling, He Shoupu in Institute of Cotton Research of Chinese Academy of Agricultural Sciences
2012-2013	Guangxi, Guizhou, Yunnan province	26 <i>G. arboreum</i> L. and 105 <i>G. barbadense</i> L.	Sun Junlin, Jia Yinhua, He Shoupu, Pan Zaoe et al in Institute of Cotton Research of Chinese Academy of Agricultural Sciences

Table 1. Cotton germplasm exploration in China during the past years

Years	Locations	Acquirement	Explorer and institute
1984	Mexico		Hu Shaoan in Institute of Cotton Research of Chinese Academy of Agricultural Sciences, Chen Zhongfang in Institute of industrial Crops, Jiangsu Academy of Agricultural Sciences
1979-1993	Mexico, including Sonora, Sinoloba, Baja California Sur, Colima, Yucatan, Michoacan, Colima	243 wild species and Landraces, including race <i>punctatum</i> , race <i>mexicanum</i> , race <i>richmondii</i> , race <i>latifolium</i> , race <i>morilli</i> , race <i>mariegalante</i> , race <i>parmeri</i>	Shen Duanzhuang, Huang Junqi, Xiao Songhua, Zhou Baoliang et al in Institute of industrial Crops, Jiangsu Academy of Agricultural Sciences
2011-2013	Hawaii, USA	20 <i>G. tomentosum</i> Nuttall ex Seemann	Wang Kunbo, Liu Fang, Zhou Zhongli in Institute of Cotton Research of Chinese Academy of Agricultural Sciences
2013	Brazil		Wang Kunbo, Liu Fang in Institute of Cotton Research of Chinese Academy of Agricultural Sciences
2013	Australia		Wang Kunbo Liu Fang in Institute of Cotton Research of Chinese Academy of Agricultural Sciences

Table 2. Cotton germplasm exploration in cotton origination place during the past years

Collections obtained from foreign countries are showed in Table 2 and Table 3. The earlier introduction of germplasm of Upland cotton were Deltapine lines imported from USA which were the main varieties grown in China in 1950s [15-17]. Following, Acala and Stonville lines were also introduced to China which helped to enhance new cultivars by the native breeders based on these materials. From 1980 to 1997, Ma Jiazhang, Xiang Shikang, and Liu Guoqiang imported genetic stocks, PD lines and *G. barbadense* separately. The Soviet Union was also one of the countries that provided their varieties and lines including the early mature and resistant Upland cottons, short branch Sea island cottons and various hybrid lines. Recently, many accessions were exchanged between China and other countries, including Russia and Uzbekistan. The expeditions to the origin sites were mainly made in Mexico, Brazil and Australia. Till now, 2,236 cotton accessions including 2,024 *G. hirsutum* L., 198 *G. barbadense* L. and 14 *G. arboretum* L were introduced from 53 foreign countries in the last 100 years. The top five countries with a large number of introductions were United States of America, Russia, Australia, Pakistan and Egypt.

Years	Collection Issues
1977	Import 9 wild species from Czechoslovakia Prague tropical subtropical Agricultural Research Institute of Kasetsart University, including <i>G. australe</i> Mueller, <i>G. trilobum</i> (DC.) Skovsted, <i>G. tomentosum</i> , <i>G. bickii</i> , <i>G. sturtianum</i> , <i>G. davidsonii</i> , <i>G. klotzschianum</i> , <i>G. davidsonii</i> , <i>G. herbaceum</i> var <i>africanum</i>
1980	Import 50 genetic stocks from USA, and 7 <i>G. hirsutum</i> landraces from Mexico
1982	Import 21 wild species from USA, including <i>G. nandewarensis</i> Derera, <i>G. capitata-viridis</i> Mauer, <i>G. armourianum</i> Kear, <i>G. gossypoides</i> (Ulbr) Standl, <i>G. longicalyx</i> J.B.Hutch & Lee, <i>G. triphyllum</i> (Harv, Sond) Hochr, <i>G. incanum</i> (Schwartz) Hillc, <i>G. aridum</i> (Rose&Standley) Skovsted, 3 wild species from French including <i>G. turneri</i> Fryx, <i>G. laxum</i> Phillips, <i>G. nelsonii</i> Fryx.
1983	Import 27 PD lines from USA, and 33 lines from French
1984	Import 6 wild species from Mexico, including <i>G. aridum</i> and <i>G. laxum</i>
1986	Import 38 lines from Pakistan, including 6 wild species and 7 <i>G. arboreum</i> L.
1986	Import 6 wild species from The Commonwealth Scientific and Industrial Research Organisation in Australia, including <i>G. cunninghamii</i> Todaro, <i>G. costulatum</i> Todaro, <i>G. populifolium</i> (Bentham) Mueller ex Todaro
1987	Import 79 GP lines and 3 wild species including <i>G. bickii</i> , <i>G. triphyllum</i> , <i>G. anomalum</i> , and 11 lines from Pakistan
1989	Import 3 wild species from Cote d'Ivoire, including <i>G. incanum</i> (Schwartz) Hillc, <i>G. turneri</i> Fryx, <i>G. mustelinum</i> Miers & Watt
1990	Import 52 lines, including 3 wild species
1991	Import 55 lines from USA, 8 lines from French, 5 line from Egypt, 1 line from Burma, 1 line from Barbados
1992	Import 94 lines from Australia, including 16 wild species
1990-1995	Import 8 <i>G. hirsutum</i> landraces and 4 wild species from Mexico including <i>G. schwendimanii</i> Fryxell)
1999	Import 35 lines from Mexico
2001	Import 36 MAR lines from USA
2006	Import 418 <i>G. hirsutum</i> landraces from USA
2008	Import 100 lines from Uzbekistan
2009-2013	Import 990 lines from Russia

Table 3. Cotton germplasm importing and collection from foreign country

3. Status of cotton germplasm preservation in China

Funding for maintenance of the collections was provided by the Chinese government under the National Key Technology Research and Development Program, and the National Key Basic Research and Development Program, Funds primarily provided by the National Science Foundation of China, the Ministry of Science and Technology, and the Ministry of Agriculture. There are five full-time staff members assigned to the maintenance and conservation of the germplasm collection. The total collection consists of 8,868 accessions that represent cultivated species (*G. arboreum*, *G. herbaceum*, *G. hirsutum*, *G. barbadense*) and 32 wild species (Table 4 and

Table 5). In these collections, most of the accessions were originated from all China, and the other accessions were imported from 52 countries (Table 6).

Species	Total	Catalogued	Cataloguing
<i>G. hirsutum</i> L.	7402	6305	1057
<i>G. barbadence</i> L.	633	532	101
<i>G. arboreum</i> L.	433	350	83
<i>G. herbaceum</i> L.	18	15	3
<i>G. hirsutum</i> landraces	350	350	
Wild species	32	15	17
Total	8868	7567	1261

Table 4. Cotton germplasm in the medium-term germplasm bank

7,221 accessions were preserved for long-term in Beijing at -18°C under 57% relative humidity. The collection in Anyang, considered the working collection, contains 8,868 accessions and is maintained at 0°C under 50% relative humidity. The Hainan Island nursery site maintained an *in vivo* collection of 391 wild species and *G. hirsutum* landrace stock accessions in a garden under natural field conditions [18-19]. Over time, the Chinese collection has grown through international germplasm exchange and expeditions. The germplasm expeditions covered most of southern China (Hainan Island, Guangxi, and Guangzhou) and the mountainous areas of Yunnan and west Sichuan provinces. *G. hirsutum* landrace germplasm has also been collected from southern Mexico.

The seeds of germplasm accessions in the working collection were renewed according to individual accession germination rates. When germination rates fell under 65%, or the weight of individual accession was no more than 150g, these accessions would be increased and renewed for the maintaining of the viability, genetic integrity and releasing for the research. 50-100 seedlings of each individual should be kept in the field for the harvest of enough healthy seeds. The field was chose according to the environments of origins of the accessions for propagation. The *G. hirsutum* L. has been propagated in Anyang, Nanjing, Kuche, and Liaoyang, in the past years. The *G. barbadence* L. has been grown in Kuche or Turpan of Xinjiang because of low rainfall and high temperature weather. The lines sensitive to the Verticillium wilt and Fusarium wilt were grown in disease free field. Some lines that were photoperiod and temperature sensitive were grown in Sanya of Hainan province. The main agronomic traits were checked according to the database descriptors in the germplasm bank, and the mixed seedlings were weeded out for keeping the purity of the accessions and the true accuracy of the database. Cotton germplasm preserved in the local institutes last century were renewed for enough seeds with purity and vitality. Every year more than 300 accessions were renewed for better seed vitality (Table 7).

Species	Origin
<i>G. nandewarensis</i> Derera	Nandeware, New South Wales, Australia
<i>G. sturtianum</i> Willis	Centre and South Australia
<i>G. robinsonii</i> Muell	West Australia
<i>G. costulatum</i> Tod	Kimberley, Australia
<i>G. cunninghamii</i> Tod	North Australia
<i>G. populifolium</i> (Benth) Muell ex Tod	Kimberley, Australia
<i>G. australe</i> Muell	Centre and North Australia
<i>G. bickii</i> Prokhanov	Centre Australia
<i>G. nelsonii</i> Fryx	Centre Australia
<i>G. thurberi</i> Tod	Arizona, USA; Sonora and Warburg, Mexico
<i>G. trilobum</i> Skov	Mexico
<i>G. davidsonii</i> Kell	Baja California, Mexico
<i>G. klotzschianum</i> Anderss	Galapagos islands
<i>G. armourianum</i> Kearn	San Marcos and Baja California, Mexico
<i>G. harknessii</i> Brandg	Baja California, Mexico
<i>G. turneri</i> Fryx	Sonora, Mexico
<i>G. gossypoides</i> (Ulbr) Standl	Warburg, Mexico
<i>G. aridum</i> (Rose & Standley) Skov	Mexico
<i>G. schwendimanii</i> Fryx	Mexico
<i>G. lobatum</i> Gentry	Mexico
<i>G. laxum</i> Phillips	Guerrero, Mexico
<i>G. raimondii</i> Ulbr	Centre and West Peru
<i>G. anomalum</i> subsp. <i>senarensis</i> (Wawra & Peyr) Vollesen	Niger, Chad, Sudan, Ethiopia
<i>G. capitis-viridis</i> Mauer	cape verde islands
<i>G. incanum</i> (Schwartz) Hillc	Yemen
<i>G. somalense</i> (Gürke) J.B.Hutch	Sudan, Ethiopia, Somalia, Kenya
<i>G. stocksii</i> Mast in Hook	Somalia, Oman, Pakistan
<i>G. longicalyx</i> J.B.Hutch & Lee	Sudan, Uganda, Tanzania
<i>G. triphyllum</i> (Harv, Sond) Hochr	Angola, Botswana, Namibia
<i>G. tomentosum</i> Nutt & Seem	Hawaii
<i>G. mustelinum</i> Miers & Watt	Brazil
<i>G. darwinii</i> Watt	Galapagos islands

Table 5. Wild species in the germplasm bank in China

Origin	<i>G. hirsutum</i> L.	<i>G. barbadence</i> L.	<i>G. arboreum</i> L.	<i>G. herbaceum</i> L.	Origin	<i>G. hirsutum</i> L.	<i>G. barbadence</i> L.	<i>G. arboreum</i> L.	<i>G. herbaceum</i> L.
Afghanistan	1				Kenya	5			
Albania	1	3			Mali	2			
Angola	1				Mexico	17	1		
Argentina	7				Nepal	5			
Australia	154				Nicaragua	1			
Brazil	4				Nigeria	3			
Bulgarian	14				North Korea	3			
Burkina Faso	2				Pakistan	99		1	
Burma	1				Peru	1			
Burundi	1				Portuguesa	1			
Cambodia	1				Somalia	2			
Cote d'Ivoire	10				Soviet Union	355	135		
Canada		1			Spain	3			
Chad	1				Sudan	8	1		
Cameroon	2				Sweden	1			
Cuba	4				Syria	7	3		
Czech	1				Tanzania	5			
Egypt	1	41			Turkey	6			
France	29				Uganda	11			
Ghana	1				USA	1200	13	11	
Greece	6				Uzbekistan	11			
Hungary	1				Vietnam	5			
India	15		1		Yemen	1			
Indonesia	2				Yugoslavia	2			
Iran	1				Zambia	1			
Japan	4		1		Other	4			

Table 6. The Collections from different countries in the medium-term germplasm bank

Years	2001	2002	2003	2004	2005	2006	2007	2008
Accession Number	997	1058	574	700	586	1336	404	400

Table 7. Seed propagation of cotton germplasm in recent years

4. Evaluation of cotton germplasm in China

For the utilization of the cotton germplasm, all the accessions in the germplasm bank were evaluated with different method, including the main agronomic traits and mutant beneficial genes. We took charge of some national projects such as “*Propagation, Identification, evaluation and utilization of cotton germplasm*”, “*Construction molecular map of cotton germplasm and molecular marker for the genes with important traits*”, “*Germplasm enhancement and utilization of cotton*” etc., for elite germplasm evaluation. All these projects were in cooperative with provincial cotton research institutes and national universities. We are seeking to have some chances to make international cooperation on collection and evaluation.

4.1. Evaluation of agronomic traits

Germplasm accessions were evaluated by field investigation according to “*Descriptors and Data Standards for Cotton Germplasm*” utilizing the IPGRI description standards for cotton [20]. Sixty-six agro-economic characters were evaluated, including 41 morphological and agronomic characters, 13 fiber quality, 4 seed quality, 8 disease, insect, multi-adversity resistance characters among 6372 accessions. The main characters were early maturity, mid-maturity, late maturity, clustering of bolls, dwarfness, pubescence, naked seeds, big boll, high lint percentage, long fiber, strong fiber, high oil content, high protein content, low gossypol content, high gossypol content, high Fusarium wilt resistance, high Verticillium wilt resistance, root disease resistance, aphids resistance, bollworm resistance, red pink boll worm resistance, spider resistance, draught resistance, salinity resistance, humidity resistance, cold resistance, color of fiber, male sterility, and genetic markers (Table 8). The Chinese Crop Germplasm Information System (CGRIS) served as a central repository for all germplasm accession characterization and evaluation information for China and was freely available at <http://icgr.caas.net.cn/cgrisintroduction.html> (verified on July 17th 2009). Cotton germplasm evaluation and characterization data was first stored on an offline, internal database and then periodically deposited onto the CGRIS online database.

The evaluation of environment adaptation was also carried out in the past years. All germplasms from the foreign origins were divided into three types based on the latitude, including tropical type, subtropical type and temperate zone type, and were grown in Lujiang in Yunan province, Nanjing in Jiangsu province, Yuncheng in Shanxi province, Kuerle in Xinjiang province. It was found that the germplasm from USA were adaptable in Yellow River area and Yangtze River area because of the similar latitude and weather, and the germplasms from

Years	Fiber quality	Fusarium wilt	Verticillium wilt	Drought	Salt	Boll -worm	Aphid	Pink worm	Tetranychid mite
1957-1990	2943	2344	2345	2844	2888	365	364	359	1
1991-2000	1242	888	897	1053	1064	337	356	337	346
2001-2005	491	470	485	477	484	198	202	208	207
2006-2009	501	416	500	487	500	284	499	500	498

Table 8. Evaluation of the main traits in cotton during the past years

Central Asia grew very well in Xinjiang province, but these from Brazil and Africa produce only in Yunan. This provided a guidance for the further introduction germplasm in different area. Some better fiber quality lines were screened after the evaluation in different environments, such as Chuan77-1, Acala1517-70, Acala1517-77, LineF, upland, FJA, Zaoshuchangrong7, ITMA96497, Hopical. Now Nanjing, Anyang, Kuerle, Kuche and Shijiazhuang were chosen as the appointed locations for the environment adaption evaluation of the cotton germplasm in China.

The *G. arboreum* L in China was selected to evaluate for special agronomy traits, such the fiber quality, abiotic resistance, specific follower color and leaf. Some important traits were classified into different types including oil and protein contents in the seeds, fiber fineness, and boll maturity based on the evaluation of all the germplasm.

4.2. Evaluation with molecular method

The genetic diversity of main sources germplasm of Upland cotton were studied on the basis of simple sequence repeat (SSR) markers. These germplasm of Upland cotton included the cultivars bred in different periods and grown in different ecological areas, the lines transferred with genes and exotic DNA, and the introgression lines from interspecific hybridization [13]. Meanwhile, SSR fingerprinting analysis were set up to indentify the elite germplasm with high yield and good fiber quality, and QTLs mapping were carried out to screen the markers linked or associated with the beneficial genes [21-24]. Those studies presented an overview of the genetic diversity of the cotton germplasm in China, and provided a guideline for breeders to develop new cultivars efficiently.

4.2.1. Evaluation of introgression *G. hirsutum* lines

The introgression *G. hirsutum* lines were created by different institutes and collected in the germplasm bank in the past years, which possessed some important beneficial genes from the wild species. Our research showed that there were specific SSR loci among the different exotic *G. hisutum*. Around 45.2% of 155 interspecific lines possessed specific SSR loci [13]. The percentage of the lines with exotic specific SSR loci among the exotic *G. hirsutum* which may be transferred from *G. barbadense* and *G. thurberi* was highest, reached 62.1% and 71.1% respectively. The second was those with the specific SSR loci of *G. anomalum* and *G. bickii* etc.,

reached 57.1% and 42.9% respectively (Table 9). Because the numbers of exotic gene transformation were different, the specific SSR loci detected were very different. The *G. hirsutum* with exotic *G. barbadense* genes possessed 1-4 specific SSR loci, and those with *G. thurberi* had 1-5 specific loci. Through analysis of the agricultural traits of the introgression lines with specific SSR loci, the correlation between specific SSR loci and the main elite traits of the introgression lines was found. The results showed that the more specific SSR loci of exotic species, the better the fiber length and strength, the more tolerance of drought stress etc., and the elite strains with resistance to Verticillium wilt maybe selected from the germplasm with specific SSR loci (Table 10). This indicated that there were desirable markers and genes related to high fiber quality, tolerance to draught, resistance to Verticillium wilt etc. among these exotic *G. hirsutum* lines. For example, the variety Shenmian 5 in *G. hirsutum* with exotic genes of *G. barbadense* had 5 specific SSR loci. This caused its high fiber quality with length 34.1 mm and strength 24.3 Cn/tex, which indicated the high fiber quality of *G. barbadense* may be transferred into Shenmian 5. Similarly, the character of resistance to Verticillium wilt of *G. hirsutum* variety Shenmian 718 maybe resulted from *G. barbadense* since it had 2 specific SSR loci of *G. barbadense*. The high fiber quality traits of *G. hirsutum* variety Acala SJC-1, FJA and J line with consanguinity of *G. thurberi* maybe also resulted from potential high quality genes of *G. thurberi*. Moreover, the similar molecular and agronomic characters of exotic *G. hirsutum* of *G. barbadense* and *G. thurberi* indicated *G. barbadense* may be originated from *G. thurberi* [13].

Exotic species	No. of accessions	No. of specific loci	Percentage of lines with specific loci(%)	Average specific loci	Range of specific loci
<i>G. barbadense</i>	29	8	62.1	2.6	1-4
<i>G. arboreum</i>	21	2	4.8	2.0	2
<i>G. thurberi</i>	45	15	71.1	1.7	1-5
<i>G. sturtianum</i>	12	2	25.0	2.0	2
<i>G. bickii</i>	7	1	42.9	1.0	1
<i>G. anomalum</i>	7	3	57.1	1.8	1-3
<i>G. hirsutum</i> race <i>mexicanum</i>	5	1	20.0	1.0	1
<i>G. raimondii</i>	5	2	40.0	1.0	1
Total introgression lines	155	25	45.2	1.8	1-5

Table 9. The specific SSR loci of introgression genes among some exotic *G. hirsutum*

For confirming the detail information of each accession, the database of fingerprints was set up to describe and distinguish some lines. "Decimal string code" was given to each individual based on the fingerprints after running gel with SSR markers, and then the information of the code as an identification number was integrated with the existent database. This helped to keep the uniformity of the accessions in the germplasm bank and the releasing lines. Till now

	Drought tolerance (%)	Salt tolerance (%)	Verticillium Wilt tolerance (%)	Fusarium Wilt Resistance (%)	Bollworm resistance (%)
Specific SSR loci	0.521**	-0.051	-0.100	-0.068	0.026
Lines with specific loci(%)	-0.029	-0.439*	0.475*	-0.288	-0.494**
	Length (mm)	Uniformity (%)	Strength (cN/tex)	Elongation (%)	Micronaire
Specific SSR loci	0.185	0.020	0.085	-0.009	-0.139
Lines with specific loci(%)	0.215	0.192	0.438*	0.319	0.100

* showed significance at 5%, ** showed significance at 1%

Table 10. Correlation coefficient between SSR loci and fiber traits and stress resistant traits

330 cultivars have been assigned with the “Decimal string code”, and more lines will be done in the future [25].

4.2.2. Identification of the genes or QTLs in the specific germplasm

For further finding and using the potential beneficial genes, QTLs mapping and cloning of genes were carried out using the mutant lines that were developed through physical and biologic methods or/and collected from other institutes. A new fiber developmental mutant *G. hirsutum* line GZnn with fuzzless-seed was analyzed by traditional genetics methods and SSR marker analysis. It was found that GZnn possessed a qualitative trait of naked seed that was controlled by one recessive gene. Using SSR marker technology, we located this fuzzless-gene on chromosome 10 and named it n_4 , which was different from N_1 (on chromosome 12), n_2 (on chromosome 26) and n_3 (found in USA in recent years). This gene was closely linked with SSR marker S1495-120. Some other novel fiber quality QTLs were also detected using genetic population of the interspecific crossed line J02-508 which was segregated for fiber length and fiber strength. The fiber length of line Su7235 was found to be controlled by one major gene and many minor genes. A dwarf gene which was thought to be caused by the absence of gibberellin (GA) and recovered by spraying GA in the seedling stage, was also found and mapped[22]. Other genes about the big boll, high lint percent, brown fiber, resistance to *Verticillium* wilt, and glandless in the seedling [21-24]. Some important genes controlling the drought and salt tolerance, yield, fiber quality and boll opening stage, were screened with the method of association mapping using a germplasm resources.

5. Germplasm release and utilization

The purpose of the collection and evaluation of cotton germplasm is to release and utilization for the improvement of breeding based on the screening of the beneficial genes. The procedures

were made to fit for the normal release and utilization for the research. Since recorded files, a large number accessions have been released and helped to promote the yield, quality and resistance in the development of cotton breeding in China.

5.1. Germplasm release

Formal germplasm seed requests within China are needed to provide to the Cotton Research Institute of the Chinese Academy of Agricultural Sciences. A Material Transfer Agreement is required to distribute seeds of requested germplasm. Seed requests from outside of China require a complex procedure for approval. Cotton characterization and evaluation database is linked with Chinese Germplasm Resource System (CGRIS: <http://icgr.caas.net.cn>). More than 11241 accessions cotton germplasm have been released for the customers in China and a few foreign counties till 2006 (Table 11). Since 2007, different accessions have been grown in the field every year and have been arranged for demonstration in the boll opening stage for the selection and utilization of the researchers. Through this way, the researchers could visually recognize and know the lines in the germplasm bank, and decide what they want to choose for the breeding and other research. Therefore, the numbers of accessions released in recent years which were selected by the researchers themselves increased dramatically every year.

Province/ Country	Accession number	Number of organizations	Province/ Country	Accession number	Number of organizations
Henan	2525	27	Shanghai	20	4
Xinjiang	1803	21	Tianjin	11	3
Jiangsu	981	22	Jilin	4	1
Hebei	863	24	Xizang	3	1
Beijing	862	15	Guangdong	1	1
Shandong	573	18	Yunnan	1	1
Hubei	542	15	USA	204	
Jiangxi	422	9	Brazil	52	
Hunan	356	11	Russia	50	
Sichuan	330	5	Vietnam	18	
Shanxi	314	8	Australia	12	
Zhejiang	257	4	Ethiopia	10	
Liaoning	256	6	Pakistan	15	
Anhui	122	16	Marcello	6	
Shanxi	101	5	Kazakhstan	3	
Gansu	52	5	India	1	
Hainan	28	4	Others	190	
Chongqin	25	2	Total	11241	

Table 11. The accessions released in the different areas during the past years

5.2. Germplasm utilization

5.2.1. Utilization of basic germplasm

All these accessions shared with the researchers have helped to develop some import cultivars. Some basic accessions have made important contribution to the improvement of cotton breeding, which could be summarized in three stages in the development of cotton germplasm. In the first stage, the germplasm imported from USA, such as TX 53, Stoneville 4, Stoneville 4B, Stoneville 2B, Coker 100, D.P.L 14, D.P.L 15, and Empire, were the main basic lines to promote the breeding level in China [15-17,26-27]. More than one thousand new lines were bred based on these basic lines. Guannong No.1 is the typical cultivar that was bred using the basic line King, and also was used as new basic line for the new cultivar breeding. In the second stage, the cultivars and germplasm selected by the Chinese breeders, such as Yishuhong, Jingmian 1, Ganmian 1, Xuzhou 209, Xuzhou 1818, Dongting 1, Zhongmiansuo 2, Zhongmiansuo 3, Zhongmiansuo 4, Zhongmiansuo 7, Jiangsumian 1, Jiangsumian 2 and Jiangsumian 3, became the new basic lines, which were commercialized and grown in larger areas. Some lines with resistant genes also were used as the basic lines, such as Liaomian 1, Liaomian 3, Shanmian 3, Shanmian 4, Shanmian 5, 52-128, 57-681, and D.P.L 16 from USA, as well as 24-21 and KK1543 from the Soviet Union. New cultivars that were created using these basic lines, replaced the varieties imported from foreign countries in commercialization. In the third stage, the basic lines with the resistant to the disease, early maturity, high lint percentage and male sterility, were used that resulted in the resistant lines 86-1 and Zhongmiansuo 12, early mature lines Heishan mian 1 and Zhongmiansuo 10, high lint percent and heterotic line Jimian 1, and male sterile line Dong A. The glandless lines such as Lambright GL-5 and Mcnair 210 introduced from USA, helped to create new type of cultivars in China. The basic germplasm lines of *G. barbadense* were fewer than the *G. hirsutum*. Only seven lines were most frequently-used in the breeding of *G. barbadense*, such as C6022, 8763 I and 9122 i from the Soviet Union, Menoufi from Egypt, Xinhaimian, Junhai and Changrong 3 from China.

Some important cultivars have been bred after the basic lines were introduced to the breeders in China. Zhongmiansuo 12 was from the crossing of Uganda 3 and Xingtai 6871, which was the first high yielding cultivars that was resistant to Fusarium wilt and was ever grown in nearly 50% cotton area in China during 1992-1996. Lines 52-128, 57-681-52-128 and 57-681 were selected from D.P.L 15 and Texas 531 bred by Sichuan cotton institute, and became the important lines that provided the gene source of resistance to Fusarium wilt. In China, Liaomian 1 and Liaomian 2 were the pioneer varieties with Verticillium wilt. Resistance. Two varieties of Zhong 8004 and Zhong 8010 resistant to Verticillium wilt were produced. Lines Shan 1155 and Zhong 3474 with resistance to Fusarium and Verticillium wilt provided resistant genes for seven varieties. In the 1990s, almost 111 varieties had the resistant genes of Verticillium wilt from the lines 86-6, Chuan 737, Chuan 2802, Shan 2234, Shan 6192, Chuanmian 239, Huai 910 and Chuanmian 243.

5.2.2. Utilization of germplasm with beneficial traits and genes

Germplasm with beneficial agronomic traits were utilized in the past year for purpose of the good quality, high yield, and multiple resistance in the cotton breeding. Nine PD lines with good fiber quality, such as PD 4548 and PD 2164, have been the origin of eleven new cultivars including Emian 21, Lumian 14, and Wanmian 10. The new varieties Xinluzao 6, Xinluzao9 and Xiangzamian 9 came from the contribution of line Bellsiro with good fiber quality [24, 26]. Lumian 9 had the pedigree of Acala. In the field of resistant research, Simian 3 that was important and popular in commercialization, was developed by importing the Fusarium wilt resistant genes from the line Yankang 76-75. Also Liaomian 5 was the origin of Verticillium wilt resistant gene and contributed to the breeding of six new varieties including Zhongmiansuo 20 and Lumian 7. Chuanmian 109 that possessed the gene of resistance to aphid, helped to create the cultivars Chuanmian 109-1 and Jingmian 1 with the resistant to the aphid. In recent years, more and more varieties were bred through composing different kinds of powerful genes from the special germplasm lines. For example, Chuang 075 was from the lines Zhongmiansuo 12, Yumian 19 and Zhongmiansuo 21 with Verticillium wilt resistance. Zhongmiansuo 44 developed from the line Zhong 2369 with Fusarium wilt resistance, the line Tamcot CD3Hal with drought tolerance and the line Zhongmiansuo 17 with high yield. Zhongmiansuo 49 derived from the early mature line Jing 444 and the line Zhongmiansuo 35 with salt tolerance and high yield. Zhongmiansuo 48 with good fiber quality, big boll and high yield, was from the big boll line Su9108.

5.2.3. Utilization of special germplasm with specific traits

Some special cotton germplasm showed different characteristics to the standard lines in the traits of leaf shape, flower color, bract shape, absence of nectarines, fiber color etc. The first special trait used in the breeding was okra leaf, which reduced the vegetative growth for keeping well light ventilation, and was resistant to *Syllepte derogata Fabricius*. The varieties with okra leaf were very popular in the cotton growing areas. The oldest cultivar with okra leaf used in china was Okra leaf TX 8207 selected from normal leaf TX 8207, which was the important variety before 1958. The second famous cultivar with okra leaf was Biaoza A1, a hybrid commercialized in Xinjiang. The virescent trait was also used in the breeding for indentifying the false hybrid in the seedling stage. Frego-bract providing the resistance to the bollworm, pink bollworm and boll weevil through the narrow and twisted bract, helped to create the new cultivar Zhongmiansuo 33. The absence of nectary in the leaf and bract could reduce the attraction to insects, and was used in the breeding of morphological resistance to insects such as Zhong 5909 and Ji U82-3[28]. The glandless trait attracted the researchers when it was found to be lower poisonous to the health of people for the oil and protein consumption. Many varieties with glandless such as Zhong 13 and Yumian 6 have been selected in the past years and were grown in some special areas. The cultivars with brown and green fiber were also created. The first two colored cotton cultivars in China were Zhongxu 1 and LvXu 1, which genes were transferred from the wild cotton. In recent years, more and more colored cotton cultivars were bred by different researchers, such as the brown fiber cultivars Zhong 263,

Zhong 163, Zhongmiansuo 51, Xincai 3, Xiangcaimian and Zhongmiansuo 81, and green fiber cultivars Lv 4560 and Zhongmiansuo 82.

5.2.4. Utilization of wild species

Wild species possess many beneficial genes that have been swept in the domestication of cotton. Researchers in China started to study and utilize the wild species very early. The most famous variety was Shiyuan 321 from the hybridization of *G.thurberi*, *G.barbadense* and *G.hirsutum* by the institute of Genetic and Developmental Biology, Chinese Academy of Sciences(CAS) and Shijiazhuang Academy of Agricultural Sciences. Jinmian 2 was the second commercialized variety from the *G.thurberi*, *G.anomalum* and *G.hirsutum*, which possessed the traits such as resistance to Verticillium wilt, Fusarium wilt and bollworm, early maturity, high yield, and good fiber quality. Qinyuan 4 was commercialized with the multiple cross of *G. sturtianum* and two *G. hirsutum* (Zhongmiansuo 12 and Acala SJ-1), which is resistant to Fusarium wilt and tolerant to drought. Other lines were also created by different institutes in China, such as the line HST2 from the hybridization of *G. sturtianum* and *G. hirsutum* with the resistant to Verticillium wilt, Fusarium wilt, pinkworm and drought, line HAS from *G. sturtianum*, *G. hirsutum*, and *G. arboretum*, line 92-26-2 from *G. hirsutum* and *G. anomalum*, line 92-48-1 from *G. thurberi*, *G. barbadense* and *G. hirsutum*, line BZ201 from *G. thurberi*, *G. hirsutum* and *G. hirsutum* landrace, line BZ401 from *G. hirsutum* and *G. anomalum*, line JSCG7235 from *G. hirsutum* and *G. anomalum* with long fiber length and strong fiber strength.

6. Future perspectives and suggestions

In future, further research need to be carried out based on the current result of cotton germplasm in China for providing the development of cotton breeding. First, expeditions to the origin sites should be emphasized in the following stage of collection, especially expeditions in the origin sites of wild species and landraces. Exchange with the countries that had special and more diversity germplasm should be continued for enriching the diversity of cotton in China. Second, the method of evaluation should be renewed, and new methods such as the association analysis with new generation sequencing technology should be used in the procedure of screening of important germplasm carrying beneficial genes. The genes with drought and salt tolerance that are required urgently in North-west China cotton production suffering from water deficiency and salinity, should be identified accurately with high throughput methods. Third, new lines should be created through comprehensive techniques using the current germplasm resources. One novel line could significantly change the reputation of cotton breeding. Forth, high efficient database should be set up for recording all the information of the cotton germplasm, including the agronomic traits and molecular data.

Author details

Yinghua Jia*, Junling Sun and Xiongming Du

State Key Laboratory of Cotton Biology/ Institute of Cotton Research of Chinese Academy of Agricultural Sciences (ICR, CAAS), Anyang, Henan, China

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