Gossypium barbadense: An Approach for in Situ Conservation in Cerrado, Brazil

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Abstract

Abandonment of planting of *Gossypium barbadense* has endangered its existence. The objective was to determine the characteristic of the maintenance of *Gossypium barbadense* in the Central-West Region of Brazil, with the aim to foster the conservation of the species. Expeditions were conducted in 2014-2015 in Southeast Goiás, where cotton collection has not been reported before. Data from previous collections in Goiás, Mato Grosso do Sul and Distrito Federal available in Albrana database were considered this study. In the Central-West Region of Brazil, 466 accesses of *G. barbadense* were recorded, found most frequently in backyards (91.4%), but also spontaneous plants (7.5%), farm boundary (0.8%) and commercial farming (0.2%) have also been found. The main use indicated by VDU was as medicinal plant (0.66), therefore this is the main reason for in situ preservation. However, this cultural habit has been abandoned, causing uncertainty about the continuity of preservation. There was presence of lint almost only in seeds of plants of the variety *brasiliense*. The most common fiber color was white, not cream color, suggesting that plants are derived from commercial cultivars used in the past. Hence, until 2015, the conservation relies on single plants almost exclusively in backyards for medicinal use, and there are no crops of this type of cotton for commercial purpose. The exploitation of the possible use both of the fiber as a medicinal plant would foster ex situ and in situ preservation of this important genetic resource.

Keywords: arboreal cotton plant, Gossypium barbadense, in situconservation, pre-breeding

1. Introduction

Cotton is the most important natural fiber in the world. It belongs to the Malvaceae family and the genus *Gossypium* (Fryxell, 1979). The *Gossypium* genus includes about 50 different species distributed in the Asian, American, Australian and African continents (Freire et al., 2002). This wide distribution corroborates with the high morphological and cytological diversity observed in the genus (Fryxell et al., 1992), organized in nine different genomic groups namely, eight diploid (2n = 2x = 26) and one allotetraploid (2n = 4x = 52) (Endrizzi et al., 1985; Grover et al., 2012). In this diversity of species only four are cultivated, among these two are diploids, known as the Old World cottons (*G. herbaceum* and *G. arboreum*) and two are alotetraploids, known as the New World cottons (*G. harbadense*) (Wendel & Cronn 2003; Sousa, 2010).

In Brazil, there are three of the six existent alotetraploide species in the world: *G. hirsutum*, *G. barbadense* and *G. mustelinum* (Borém et al., 2003; Grover et al., 2015), hence ranking the country as an important center of dispersion and diversity of allotetraploid cotton. *G. mustelinum* is the only kind of native cotton of the country and distributed mainly in the northeastern semi-arid (Menezes et al., 2014). There are two botanical varieties of the cotton plant *G. barbadense* in Brazil: var. *brasiliense* e var. *barbadense* (Almeida et al., 2009). The *G. barbadense* var. *brasiliense* is known as kidney cotton because their seeds are adhered to each other in a form similar to a kidney and lint is absent. The variety *G. barbadense* var. *barbadense* called Maranhão or Quebradinho contains separated seeds and lint is also absent (Freire, 2000). Discrimination among varieties by the morphology of seed morphology may be non-accurate since it is controlled by one or two gene loci.

The species *G. barbadense* is valued for its high fiber quality, often referred as Egyptian cotton. It had millenarian use in the manufacture of ropes, fishing nets in Africa and it has been commercialized in the United States, becoming a product of great importance in other countries like Egypt, India and Sudan (Todou & Konsala, 2011). Also known as "pima cotton" in the northern hemisphere, still has great importance for the production of high quality fabrics (Sousa, 2010). This type of cotton was widely cultivated. In Brazil, it was introduced from Peru in pre-Colombian periods, and its cultivation as economic exploitation was made from the 1760s, initially in Maranhão state and then covering a larger region of Northeast Brazil for export to England (Alves, 2006). Its cultivation has been gradually replaced by upland cotton belonging to the species *G. hirsutum* (Braga Sobrinho & Lukefahr, 1983), since upland cotton presents the most productive varieties and has been improved for mechanized planting, being cthe first species responsible for all the production of the world's commercial fiber (Bertini et al., 2005).

Brazil is the fifth largest cotton producer in the world (Abrapa, 2015), and the Central-West Regions the leading producer, where about 630,800 ha have been cultivated in the 2015/16 season (Conab, 2015). Immediate conservation measures are necessary to protect the species germplasm (Maxted et al., 1997), since gene flow is not controlled (Ferment & Zanoni, 2007). Research on *G. barbadense* cotton have highlighted its importance as a primary source of genetic variability that can be exploited to improve the commercial cotton fiber quality (Wang et al., 2011) and as a source of genetic resistance to nematode (Gutiérrez et al., 2010).

The species *G. barbadense* occurs in the country as cultivars and local varieties, and is not commercially cultivated in Brazil. Itcan be found in all Brazilian states most frequently as a backyard plant (Albrana, 2016). Thus it has been suggested thatin situ conservation depends on the use of the plants by the owner, which may be applied to medicinal proposes or use of the fiber (Almeida et al., 2009). The changing cultural habits, environmental depredation, agricultural expansion of commercial cotton and policies of cotton eradication with the aim of pest control have been the main threats to the disappearance of numerous local varieties of cotton, and their genetic potential has not been examined yet (Barroso et al., 2005; Menezes et al., 2010; Menezes et al. 2015).

The prospection of Southwest Goiás which had not been explored before associated to the data available at Albrana website lead to a description of *G. barbadense* distribution in the Central-West Region of Brazil to promote genetic resources conservation strategies.

2. Method and Methods

Expeditions to collect *G. barbadense* were conducted in the 2014-2015 period in Southeast Goiás municipalities. The expeditions started from a ginnery, which name is California, located in the city of Ipameri, GO, then extending to adjacent cities. The following points of collection were defined by adopting a methodology "snowball" (Bernard, 1994) in which a first preconceived informant indicates another possible informant with the set characteristics of knowing local cotton plants, and so on, including more participants. The expeditions were carried out with the authorization of Information and Authorization System in Biodiversity (SISBIO No. 37165), so leaves or petals were collected for genotyping studies and seeds with the aim to create a germplasm collection in IF Goiano Campus-Urutaí, GO, Brazil.

Southeast Goiás has been chosen since there were no previous cotton collections there (Albrana, 2016), while surrounding areas of Central-West Region of Brazil were represented in Embrapa collection. A total of 39 plants were collected, covering the municipalities of Bela Vista, Cristianópolis, Ipameri, Palmelo, Piracanjuba, Pires do Rio, Santa Cruz and Urutaí.

Cotton plants were scrutinized following a semi-structured form comprising the geographical coordinates (latitude, longitude and altitude obtained by Global Positioning System-GPS Garmim®, ETREX 10), information about population (number of plants per collection point, species, type of population, declared seed origin, presence of petal stain, presence of fuzz, leaf color), cultural procedures (fertilization, seed storage and processing), as well as environmental and phenological information (time of flowering, plant height and age).

The cotton type was classified according to the definition proposed by Johnston et al. (2006) wherein: i) Wild is a kind of plant that occurs only in natural environments and that has not gone through selection; ii) Feral is sexual or vegetative progeny derived from selected crop genotypes, which has survived and self propagated for a long period; iii) Backyard plants are grown in backyards or gardens and the origin of seeds is from relatives, friends or neighbors; iv) Local variety is the traditional cotton grown by farmers and seeds are those harvested from their crops; v) Volunteer or spontaneous plants are those germinated without human interference, which seeds or vegetative propagules are often from domesticated genotypes, occurring within a crop field close to it. The type farm boundary, not considered by Johnston et al. (2006), was defined as cotton planting intercropped with other species, without commercial purposes.

For in situ characterization the data were tabulated and systematized in Excel. We included data of plants previously collected by Embrapa using the same form, in the same state, Goiás, as well as from the neighboring states of Mato Grosso, Mato Grosso do Sul and the Distrito Federal, which comprises the Central-West Region. This data is available in ALBRANA (www.cnpa.embrapa.br/albrana) and refers to the collections from 2004 to 2009. The 119 accesses from state of Goiás (GO) have been collected in 30 municipalities; the 62 of Distrito Federal (DF) from seven municipalities; the 177 of Mato Grosso (MT) from 15 municipalities and the 69 of Mato Grosso do Sul (MS) found in 17 municipalities.

A thematic map of cotton distribution was generated using the software DIVA-GIS (Version 7.5). The map layout was obtained in the website from Brazilian Institute of Geography and Statistics (IBGE). The Use of Diversity Value (VDU) was estimated according to the expression VDU = number of times in which a specific use has been cited divided by the total number of uses, as suggested by Donazzolo (2012) as a tool to describe how the local knowledge about the use is distributed.

3. Results and Discussion

Populations of *G. barbadense* from Southwest Goiás were kept mostly in backyard (91.4%), and volunteers (7.5%), farm boundary (0.8%) and commercial farming (0.2%) have also been found as shown in Table 1.

Considering these new data in conjunction with other collections in the Central-West Region of Brazil, a total of 466 *G. barbadense* accesses were recorded, 158 from Goiás State, 177 from MT, MS from 69 and 62 from Distrito Federal (Figure 1, Table 1). This amount represents 25.5% of all species accessions collected in Brazil available in Albrana. The collections in MT comprised Pantanal and Cerrado biomes, while in MS Cerrado only; indicating that a new expeditions should be carried out to the forest region in MT, and Atlantic Forest is also an area not explored yet (Barroso et al., 2005).



Figure 1. Distribution of *Gossypium barbadense* accesses in the Central-West related to collections between 2004 and 2015

Goias					Distrito Federal					Mato Grosso							Mato Grosso do Sul					
Kind of Population	М	Mt	0	Cw	z	Total	М	Mt	Cw	Z	Total	М	ц	0	z	Ag	Total	М	Cw	Z	Total	Sum
Backyards	32	-	4		94	131	48	7	7	8	60	151	-	5	16		170	53	-	11	65	426
Spontaneous	5			_	4	La				5	7	_			-		2			4	4	35 4
Crop-Bred variety	ī	ı														-	-					-
Boundary farm		ī										4				ī	4					4
Properties																						
Urban	27	-	4	·	78	110	8	ī			8	130	-	-	12	-	145	46	-	8	55	318
Rural	S			-	5	11	40	7	7	6	53	15		-	,		16	9	ī	-	×	88
Bulding Sit	-				28	29				-	-	7			З		S	-		S	9	41
No identified			i.		-	7						3					З					5
Border Road	-	ī	ī		2	9			ī			9	ī		7	ī	×	,	,		ī	14
Permanence (Years)																						
Até 1 year	4	ı.	7	ı	47	53	9	ı	ī	ī	9	34	ı	-	-	-	37	9	ī	-	7	103
2 a 3 years	25	-	7	1	52	81	٢	-	,	ŝ	11	101	-	-	12		115	38	ī	11	49	256
4 a 5 years	4	,	i.	ï	٢	Π	14	-	-	5	21	15	ī		-	,	16	٢	-	3	11	59
5 a 10 years	-	ī	ī	ı	6	10	21	ī	-	7	24	5	ī	ı	ī	ī	2	7	ī	ī	7	41
No identified					б	ŝ						-			ŝ		4				ī	2
Total	34	-	4	-	118	158	48	7	7	10	62	156	-	7	17	-	177	53	-	15	69	
UDV	0.22	0.01	0.03	0.01	0.75	-	0.78	0.03	0.03	0.16	-	0.88	0.01	0.01	0.1	0.01	-	0.77	0.01	0.22	-	

Table 1. Occurrence of Gossypium barbadense 466 access due to the use by State of the Central West Region, Brazil

Note. M = Medicinal; Mt = Making thread; O = Ornamental; Cw = Cotton wool; N = No use; Ag = Agricultural; CW = Central-West; UDV = Use of Diversity Value.

In all states, most of the accesseswas collected in backyards, 131 in GO, 60 in DF, 170 in MT and 65 in MS, totaling 87.1%. Most of the dooryard cotton plants were located in urban properties representing 68.2% of the collected plants. Dooryard plants in rural properties were 18.9% of the total. Only in Distrito Federal most of the plants werein rural area (53), while in the GO, MT and MS states they were in urban areas, 110, 145 and 55 plants, respectively. Results indicate anthropic dispersion even after replacing the cultivation by species of annual cotton plants.

Therefore, the backyard cultivation is the main form of in situ maintenance of *G. barbadense*, considered a spontaneous species in Brazil (Stalcup, 2000). Thisalsooccurs in Pará and Amapá (Almeida et al., 2009), Maranhão, Piauí, Ceará, Rio Grande do Norte, Paraíba (Menezes et al., 2010), as well as Bahia, Pernambuco, Alagoas, Sergipe, Roraima and Amapá states (Albrana, 2016). The conservation in backyards also occurs in Mexico (Ulloa et al., 2006). The complexity and difficulty of in situ conservation strategies lies therefore on the need for cooperation of the owners of the plants, and the possibility to foster the commitment to conservation of the species, according to the International Treaty on Plant Genetic Resources for Food and Agriculture (FAO 2016).

During the collections from 2004 to 2015, five categories of use were reconded: medicinal, making threads, ornamental, cotton wool, agricultural and also plants for which no use was reported (Table 1). The main type of use as the average Use of Diversity Value (VDU) in the CW was medicinal (0.66). The VDU of this category was prevalent in all states investigated in this study, ranging in 0.88 in MT to 0.22 in GO. The medicinal use also will be the main motivator for in situ conservation of *G. barbadense* in the North (Almeida et al., 2009) as well

as in the Northeast of Brazil (Ribeiro, 2008). This type of care with cotton plant is directly related to your use of leaves and cotton boll to produce homemade infusions (Stalcup, 2000). This use is associated with the bioactive compound called gossypol that is present in the genus, and prominently in the studied species, presenting an antimicrobial action (Yildirim-Aksoy et al., 2004); and antianaemic, with hematopoietic potential as seen in Wister rats (Muhammad et al., 2014).

Besides medicinal use, gossypol acts as a natural defense in protecting plants against pathogens (Botteger et al., 1964; Scheffler et al., 2012) and insects (Guo et al., 2013; Zhou et al., 2013). *G. barbadense* plants showed more tolerance to *Alabama argillaceae* and *Pectinophora gossypiella* compared with *G. hirsutum* (Hoffmann et al., 2013). Thus, the importance of these accesses can be better evaluated if the range of action of components such as gossypol is studied, as well as the relationship of their biotechnological potential with the diversity of the molecule (Barreiro & Bolzani, 2009). The current perspective of continuity of in situ conservation can be related to the importance or use attributed to the plant by the plant owner. The VDU values for unknown or no use was 0.31 in average 0.31, ranging from 0.10 in DF to 0.75 in GO. This category was common in all states, demonstrating chance of abandonment of conservation, associated to changes in cultural habits, caused devaluation of cotton craft products compared to commercial textile products (Almeida et al., 2009), as the decline in the use of medicinal plants.

The use of arboreal cotton for spinning of handmade fabrics, used mainly for the production of blankets, as illustrated in Figure 2, is a rare habit (Table 1). In the reports of the various expeditions carried out by Embrapa in recent years (Almeida et al., 2009; Barroso et al., 2005) commercial production was nonexistent and was reported only in one site located in the city of Cuiabá, MT. This crop was composed of a bulk of local seeds, the production of which would be sold, but the commercial activity of this type of cotton had already been abandoned in the region (Barroso et al., 2005). The commercialization of arboreal cotton is registered by IBGE (IBGE, 2015), and includes mocó cotton (*G. hirsutum* var. *marie galante*), which is predominant. It was abandoned gradually, since in the 90s 554,000 ha was cultivated, not leaving in 2014 any area for harvest, according to the IBGE.



Figure 2. Cotton fabrics handmade from the fiber of cotton *G. barbadense* the municipalities of Palmelo (a) and Pires do Rio (b), Goiás

The permanence of the plants in situ, in the evaluated period, was up to 10 years (Table 1). However, the average age of plants was 1 to 3 years, representing 67% of observed plants, with similar distributions in all states analyzed. Considering that single plants were predominant, seeds distribution is recent, obtained from neighbors and relatives of adjacent cities, similarly to the Mocó arboreal cotton from semi-arid northeast (Menezes et al., 2010).

Assessment of qualitative botanic descriptors in situ demonstrated that there is morphological variability within and between germplasm collected in the states (Figure 3). Two botanical varieties of the species *G. barbadense* were identified in all states, var. *barbadense* and var. *brasiliense*, which are recognized by the seed morphology, separated seeds or kidney seeds, respectively. The var. barbadense also known as Quebradinho was more frequent in the states of MT and MS, differing from other states (Figure 3a). The presence of the seed fuzz, observed in seeds from all states (Figure 3b), occurred almost exclusively in brasiliense variety. This fuzz confers the seeds the appearance of a velvet, unlike the lint present in upland cotton seeds, which is longer, suggesting the absence or very small interspecific flow. The same occurs for petals stain. Although it is present in

upland cotton, a low frequency has been reported for *G. barbadense*, which han been maintained, therefore suggesting that gene flow does not occur in a high frequency, with no isolation between the of cotton species (Figure 3c).

The fuzz and fibers of tetraploid cotton presents a range of colors from white to various tones of brown and its can also be slightly green (Carvalho et al., 2014). The plants collected in the Central-West Region have white as the most common fiber color, unlike indicated by Carvalho et al. (2014), that the *G. barbadense* mostly often presents cream fuzz. This indicates that they are remaining from cultivars that have been planted commercially in the 80s (Figura 3d), a time when commercial trade was focused on the export of white fiber for Europe (Borém et al., 2003).



Figure 3. Botanical descriptors of qualitative variables in *G. barbadense* accesses by State of the Center West of Brazil. (A) Seed shape, (B) Fuzz color, (C) Petal spot, and (D) Fiber color

Based on the relative frequency of categorical variables it is clear that the distribution of variation is not random, showing the influence of anthropogenic dispersion in the distribution of species. This was expected, once this type of cotton was cultivated for decades in Brazil and, even after its commercial abandonment, it is dependent on human distribution. The absence of use of the species, is the greatest threat for in situ mantainance. In the Northeast Brazil states Paraíba and Rio Grande *G. barbadense* was no longer found in collection expeditions between the years 2004-2009 (Menezes et al., 2015). Incentives for research of this medicinal use would provide scientific bases to foster its maintenance.

4. Conclusion

In situ conservation of *Gossypium barbadense* is based on single individuals, almost exclusively in backyard for medicinal use, and there are no crops of this type of cotton for commercial purpose. It is still more diverse than ex situ materials. The exploitation of the possible use both of the fiber as a medicinal plant would foster ex situ and in situ preservation.

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