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INCREASING GENETIC DIVERSITY OF COTTON BREEDING PROGRAMS IN VENEZUELA

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ABSTRACT

There is a steady increase of demands for textile crops, where cotton is the most popular natural fiber, therefore, it is necessary to exploit cotton genetic resources. In order to develop and improve crop populations, a cotton breeding program requires access to new sources of germplasm. With the objectives to evaluate the productivity of local and introduced genotypes from Syria and Brazil, and to expand the genetic base in the Venezuelan Cotton Breeding Program, 16 genotypes were evaluated at three locations in Venezuela during 2013 and 2014 using a randomized complete block design, with three replications. The traits evaluated were plant height, stem diameter, lint yield, boll weight, number of seed per boll, 100-seed weight, and fiber content. Highly significant differences ($P \leq 0.01$) were observed among genotypes, environments and their interaction for all traits, except for lint yield and number of seed per boll across environments. 'C-2955-25' showed high yield (3514.3 kg.ha^{-1}) but was unstable across environments used in the evaluation. 'L-2955-13' was stable across the environments, suggesting that it would be a good parent. All the genotypes obtained values of lint percent above of 38%. The Syrians genotypes 'Aleppo-11' and 'Aleppo-90' showed high lint yield and good agronomic performance. Genotypes identified in this study could be used as potential sources of germplasm to be introduced in future breeding programs.

Keywords: Foreign genotypes, *Gossypium hirsutum* L., lint yield, lint percent.

INRODUCTION

Upland cotton is an economically and industrially important cultivated crop in Asia and America, and it is one of the primary agricultural crops and most important fiber for natural clothing and textile manufacturing (ICAC, 2016). In Venezuela, this crop has several sectors in the productive chain providing employment in the fields, cotton ginning and textile industry; however, the cotton lint yield per unit area obtained by average is 1.15 t.ha^{-1} (FAOSTAT, 2018), far less than the potential yields. Several factors affect the productivity and fiber quality such as weeds and insect pest (O'Berry et al., 2009; Reeves et al., 2010), soil pH and variation in organic matter content (Johnson et al., 2002; Elms et al., 2001), moisture content and soil fertility (Johnson et al., 1999; Venugopalan and Pundarikakshudu, 1999), and hand-harvest (Campbell and Jones, 2005), these factors increase the production costs to satisfy the demands and new dynamics on fiber quality by the industry (Campbell et al., 2011; Bourland and Myers, 2015). The development of cotton varieties with high lint yield and excellent fiber qualities has been the major aim of breeding programs, however this is, a big challenge due to the negative association between yield and fiber quality attributes (Constable et al, 2015) by pleiotropic and linkage effects (Bradow and Danidonis, 2000). The progress in the development of superior genotypes would depend upon the nature and magnitude of genetic variation present in the population (Ashokkumar and Ravikesavan, 2011). Therefore, the reduced genetic gain in cotton yield observed on performance of cultivars released during the last three decades, is due to narrow genetic composition of upland cotton and repeated use of few cotton germplasms in major commercial cotton breeding programs (May et al., 1995; Meredith, 2000; Zhang et al., 2005). In cotton, traits related to yield components such as boll weight, boll density and lint percentage, are mainly under influence of environment (Hassan et al., 2005; Khan et al., 2009), meanwhile, fiber quality such as length and uniformity of fiber, is under genetics and environment effects (Yuan et al., 2005; Luan et al., 2008). According to Allen (2012), Gonzalez and Salas (2012) and Zakhidov et al. (2016), improved cotton is flexible in stability and adaptation under different environments. The adaptability of foreign germplasm is essential to select new genotypes to be incorporated in a breeding program, and this adaptation is achieved efficiently when the genotypes are exposed to different environments, locations and years. A successful plant breeding program is directly related to the superiority of its genetic composition (de Souza et al., 2009). A limited supply of alleles for traits of interest could be expanded by introducing novel alleles from other populations (Ragsdale and Smith, 2007) and developing new recombinants and hence increasing genetic variability. In order to increase cotton yield, different strategies are being implemented by the Instituto Nacional de Investigaciones Agrícolas (INIA, in Spanish), where the main focus is the introduction of tropical and subtropical germplasm from semi-arid climates to enlarge the genetic base in traits such as yield and fiber quality. After selection of superior genotypes, conventional and molecular breeding tools are used to development new populations and inbred lines. Therefore, the objective of this study was to evaluate the productivity of local and introduced genotypes of cotton under three different grown environments of Venezuela in order to identify the best genotypes to be introduced in the breeding program as source of new genes.

MATERIALS AND METHODS

Sixteen cotton genotypes were used in this study. Six Venezuelan genotypes (commercial varieties FA-90, La Llanera and SN-290; and three unreleased varieties C-5529-26, C-2955-25 and L-2955-13), five Syrian genotypes (Deir Azzor-22, Aleppo-33/1, Aleppo-90, Raqqa-5 and Aleppo-118), four Brazilian genotypes (BSR-Araçá, BSR-293, BSR-286 and BSR 269-Buriti) and DeltaPine-16, as commercial check. These genotypes were being grown as elite local and foreign cultivars to the INIA´s cotton breeding program. The genotypes were evaluated under rainfed conditions during 2013 and 2014 growing season (August- December) at three tropical lowland environments in Venezuela. The study sites were: Araure (9°36'51" N; 69°14'34" W), Ospino (9°13'17" N; 69°32'55" W) and Turen (9°15'48" N; 69°5'34" W) and are described in Table 1. The sites are designated as ARA13 and ARA14, OSP13 and OSP14, TUR13 and TUR14, corresponding to evaluations during 2013 and 2014 at Araure, Ospino and Turen, respectively. Entries were established using a Randomized Complete Block design with three replications at each site. The experimental unit was two row plots that were 10 m long and spaced at 0.8 m. Two seeds, 0.2 m apart, were planted per hole, and later thinned to one plant, resulting in a final population density of 62 500 plants.ha^{-1}. During land preparation at all environments, the trials received the recommended fertilization rates, 30 kg.ha^{-1} of phosphate (P_2O_5) in the form of diammonium phosphate, 25 kg.ha^{-1} of nitrogen (N) in the form of urea and 15 kg.ha^{-1} of potassium (K_2O) in the form potassium sulphate. Second and third doses of N and K_2O, (each of 15 kg.ha^{-1}, respectively) were sidedressed 25 and 35 days after the crop emergence. Cultural practices, such as weed control and insect control, including boll weevil *Anthonomus grandis* Boheman (Coleoptera: Curculionidae), were according to infestation level for the locations. In both years, pre-emergence herbicide, Prowl®400 SC (pendimethalin; 3.5 l.ha^{-1}) was applied at planting, followed by hand weeding. Data were recorded on plot basis for plant height (PH, cm) and stem diameter (SD, cm). The rows per plot were harvested by hand to determinate lint yield (LY, kg.ha^{-1}). A sample of harvested cotton was collected from each plot, to determinate yield components included boll weight (BW, g), seeds per boll (SB), 100-seed weight also known as seed index (SI, g) and lint content (LP, %). These components were determined from 50 randomly selected bolls by genotype taken prior to harvest at each replicate. Seed cotton samples were ginned on a laboratory-scale gin (TB510A, TESTEX, China) to separate lint from seeds. Analyses of variance (ANOVA) at each environment and across environments were done using generalized lineal model (GLM) in SAS 9.3 statistical software (SAS Institute, 2011). Genotypes were considered as fixed effects, and replications and blocks within replications as random effects. When significant differences were detected among entries at each location, a combined ANOVA was conducted, with genotypes and locations within each season being considered as fixed effects, and replicates within locations and blocks within replicates as random effects. Means were separated using least significance differences (LSD) at $P \leq 0.05$. The Genotype-Environment Interaction (GEI) was analyzed using Additive Main-effects and Multiplicative Interaction (AMMI) model (Crossa et al., 1990).

Table 1. Description of experimental sites. Mean data of temperature and rainfall by August to December of years 2013 and 2014.

Location	masl	Tmin (°C)	Tmax (°C)	Rainfall (mm)	Soil type
Araure	233	22.2	31.3	457.4	Entisol, Aeric Tropic Flavaquent, silt, mixed, non-acid, isohyperthermic
Ospino	164	22.4	31.0	547.1	Inceptisol; Fluvaquentic Ustropepts, sandy loam, isohyperthermic
Turen	119	22.7	31.3	509.5	Inceptisol, Fluventic Haplustepts, sandy loam, mixed, isohyperthermic

masl, meters above sea level; Tmin, minimum temperature; Tmax, maximum temperature

RESULTS AND DISCUSSION

Agronomics traits and yield components

Homogeneity of variance test indicated homogeneous error variance for each trait in the six environments and allowed for a combined across environment analysis. The genotypes exhibited highly significant differences ($P \leq 0.01$) for all traits measured across environments (Table 2). Mean squares due to environments, genotypes and their interaction showed significant differences ($P \leq 0.01$) for all traits, except for lint yield and number of seed per boll across environments where the interaction values were non-significant.

Table 2. Mean squares and significant test for yield and related agronomic traits of 16 cotton genotypes, evaluated across three environments in Venezuela, during 2013 and 2014.

Source of Variation	**df**	**LY** (kg.ha^{-1})	**BW (g)**	**LP (%)**	**SB (boll^{-1})**	**SI (g)**	**PH (cm)**	**SD (cm)**
Environment (Env)	5	28629.64	0.99 **	0.63 **	3.43	1.02 **	1187.52 **	0.04 **
Replication (Rep)	2	5595.20	0.10	1.39	0.69	1.13 **	24.50	0.01
Block (Rep x Env)	10	10087.93	0.07	1.81	0.75	0.18	60.52	0.01
Genotype	15	72326.63 **	1.00 **	17.53 **	7.69 **	2.42 **	955.66 **	0.05 **
Genotype x Env	75	201785.41 **	0.80 **	13.37 **	13.57 **	1.09 **	772.46 **	0.03 **
Error	180	22335.06	0.14	1.87	1.49	0.23	53.15	0.01
Grand Mean		3387.45	6.17	40.75	25.92	9.85	159.30	1.77
Maximum mean		3514.33	6.54	42.64	27.78	10.35	169.94	1.85
Minimum mean		3280.83	5.75	38.88	24.94	9.22	146.94	1.63
Mean of check		3435.72	6.24	40.48	25.83	10.04	167.89	1.63
R^2		0.80	0.76	0.79	0.81	0.76	0.89	0.71
SE		267.22	0.61	2.38	2.23	0.77	17.55	0.13
CV (%)		4.41	6.06	3.36	4.72	4.84	4.57	5.04

*Significant at the 0.05 probability level; **significant at the 0.01 probability level. LY, lint yield; BW, boll weight; LP, lint percent; SB, number of seed per boll; SI, seed index; PH, plant height; SD, stem diameter.

This implied that there was Genotype-Environments interaction affecting the performance of the genotype across environments, requiring further analysis of magnitude of genotype by environments using GGE biplot. Mean squares revealed the presence of variability among cotton genotypes due to their diverse origin and the environments where the trials were carried out, being rainfall the conditional variable among locations and years. In this study, coefficients of variations were low, indicating good reliability of the inferences tested and high experimental precision. In the tropics, exists wide variation among locations due to latitude, temperature, and day length, even if they are relatively closer, which makes GEI an important source of variation. Similar effects in genotypes, environments and their interaction were reported among cotton genotypes under different tropical conditions for lint yield and other agronomic traits (Gonzalez et al., 2007; Silva Filho et al., 2008; Farias et. al., 2016; Djaboutou et al., 2017). For each of the traits, the percentage of sums of squares remaining among environments, genotypes and its interactions, ranged from 75% to 95% after removing sum of square due to error and replication (Table 3).

Table 3. Portion of sums of squares (SS) attributed to environment, genotype, and genotype x environment (G x E) as a percentage of the total sums of squares remaining after removing sums of squares due to replication, block and error.

Source sums of squares	**LY** (kg.ha^{-1})	**BW (g)**	**LP (%)**	**SB (boll^{-1})**	**SI (g)**	**PH (cm)**	**SD (cm)**
Pooled error Rep	12	11	15	11	25	5	20
Remaining	88	89	85	89	75	95	80
Environment (Env)	9	33	2	15	23	41	33
Genotype	25	37	55	31	53	33	42
Genotype x Env	66	29	43	54	24	26	25

LY, lint yield; BW, boll weight; LP, lint percent; SB, number of seed per boll; SI, seed index; PH, plant height; SD, stem diameter.

The environment account for a high percentage of sums of squares remaining for plant height (41%) and stem diameter and boll weight (33%, each one). For genotype component, the traits with high values were lint percent (55%) and seed index (53%). GxE effects accounted for a relatively amount of the sum of squares remaining for all traits and ranged from 24-66%, being the variables with high values lint yield and number of seed per boll, with 66 and 54%, respectively. Mean combined values of yield and related agronomic traits are presented in Table 4. The combined analyses showed average LY of 3386.5 kg.ha^{-1}, with maximum and minimum yields of 3514.3 and 3280.8 kg.ha^{-1}, to C-2955-25 and La Llanera, respectively.

Table 4. Mean performance for yield and related agronomic traits of 16 cotton genotypes, evaluated across three environments in Venezuela, during 2013 and 2014.

	Genotype	LY† (kg.ha^{-1})	BW† (g)	LP† (%)	SB (boll^{-1})†	SI† (g)	PH† (cm)	SD† (cm)
1	DeltaPine-16	3435.72±203.14 abc‡	6.24±0.50 bcd	40.48±2.48 cde	25.83±1.86 cd	10.04±0.80 abc	167.89±13.10 a	1.63±0.12 g
2	FA-90	3429.39±286.11 abcd	6.22±0.67 bcd	39.94±2.30 ef	25.78±1.35 cd	10.35±0.69 a	147.89±16.40 gh	1.85±0.07 a
3	La Llanera	3280.83±301.45 g	6.04±0.57 def	40.51±2.24 cde	25.61±2.09 cde	10.19±0.81 ab	161.44±18.24 cd	1.79±0.10 bcde
4	SN-290	3447.50±249.30 ab	6.01±0.55 def	41.06±2.28 bcd	25.83±2.73 cd	10.30±0.49 a	162.28±10.23 bc	1.79±0.15 bcde
5	C-5529-26	3342.06±259.62 cdefg	6.12±0.62 cde	41.46±2.40 b	25.33±2.20 ed	9.51±0.95 efg	155.89±20.17 ef	1.82±0.10 ab
6	C-2955-25	3514.33±321.80 a	6.37±0.64 ab	40.97±2.35 bcd	25.39±2.52 ed	9.93±0.61 bcd	152.56±18.02 fg	1.76±0.13 cde
7	L-2955-13	3333.00±195.33 defg	6.43±0.36 ab	40.52±2.17 cde	25.83±2.26 cd	9.88±0.37 bcd	146.94±13.43 h	1.70±0.13 f
8	Deir Azzor 22	3330.39±289.89 efg	6.07±0.59 def	38.88±2.21 g	24.94±2.55 e	9.39±0.62 fg	160.94±16.26 cd	1.78±0.15 bcde
9	Aleppo-33/1	3439.22±303.43 abc	6.44±0.53 ab	42.64±1.31 a	26.00±2.59 bcd	9.91±0.79 bcd	162.28±11.83 bc	1.79±0.13 bcde
10	Aleppo-90	3425.89±292.90 abcde	5.91±0.52 efg	41.21±2.70 bcd	26.00±2.43 bcd	10.35±0.52 a	161.06±15.52 cd	1.74±0.12 ef
11	Aleppo-118	3442.72±263.70 ab	6.36±0.65 abc	41.30±2.06 bc	25.50±2.33 cde	9.22±0.61 g	154.61±24.04 ef	1.76±0.14 cde
12	Raqqa-5	3397.33±339.38 bcdef	5.75±0.60 g	40.34±2.36 de	26.78±1.83 b	9.55±0.90 ef	157.44±20.47 de	1.74±0.11 def
13	BSR-Araçá	3371.61±218.78 bcdefg	6.35±0.62 abc	40.55±1.72 cde	26.28±1.99 bc	10.14±0.60 ab	166.94±13.14 ab	1.80±0.13 abcd
14	BSR-293	3350.39±243.50 bcdefg	5.83±0.67 fg	42.48±1.83 a	27.78±1.17 a	9.71±0.55 ed	169.94±16.42 a	1.81±0.13 abc
15	BSR-286	3353.39±260.13 bcdefg	6.54±0.57 a	40.31±2.51 de	26.22±2.41 cb	9.38±0.80 fg	169.17±11.26 a	1.79±0.15 bcde
16	BSR-269-Buriti	3305.39±187.18 fg	6.01±0.52 def	39.30±2.50 fg	25.61±2.20 cde	9.77±0.76 cde	151.55±18.85 fgh	1.74±0.11 ef

†Mean±standard deviation; LY, lint yield; BW, boll weight; LP, lint percent; SB, number of seed per boll; SI, seed index; PH, plant height; SD, stem diameter. ‡Means with the same letter are not significantly different at 5% probability according to LSD.

Also, the analysis reveal that SN-290, Aleppo-119 and Aleppo-33/1 had higher LY than the commercial check. Similar results were reported by Gonzalez et al. (2005) in similar environments in Venezuela. Boll weight was directly related to the seed cotton yield. BW average was 6.17 g, where BSR-286 (6.54 g) obtained the maximum value. Also Aleppo-33/1, L-2955-13, C-2955-25, Aleppo-118 and BSR-Araçá had higher BW than the commercial check. The lint percent is the fraction of the lint separated from a seed-cotton sample by ginning. LP average was 40.76 %, where Aleppo-33/1 showed the maximum value with 42.64 %. To this trait, all the Syrian genotypes except for Deir Azzor-22, were superior to the commercial check, DeltaPine-16 (40.48%). In addition, all the genotypes obtained values of LP above 38%, which is the industry requirement in Venezuela. However, a clear trend between genotypes and environments to produce high lint percent was not observed. Similar results were reported by Murakabi et al. (2004) and Suinaga et al. (2006), they found differential responses of genotypes for LP in different environments. Lint yield and lint percent are valuable information to breeders to consider using foreign genotypes in their breeding programs (Carvalho et al., 1995; Beyer et al., 2014). The average number of number of seed per boll was 25.93. BSR-293 has the higher number of SB, with 27.78. All the foreign germplasm used in this study, with exception of BSR-269-Buriti and Deir Azzor-22, were superior to the commercial check. SI average was 9.85 g, with values of 10.35 and 9.22 g, to Aleppo-90 and Aleppo-118, respectively. Plant height average was 159.36 cm, with values of 169,94 and 149.94 cm, to BSR-293 and L-2955-13, respectively. Stem diameter was 1.85 and 1.70 cm, for FA-90 and L-2955-13, respectively, with a mean of 1.77 cm, being all the genotypes evaluated superior to DeltaPine-16. The Brazilian genotypes BRS-286, BRS-269-Buriti and BRS-293 showed higher values of plant height and stem diameter than the overall mean. These traits can be used as an indicator of yield potential (Zotz et al., 2001) and can contribute to calculation of growth rate in the flowering and boll stages to select genotypes that more effectively utilizes their energy to form cotton products (Jiang et al., 2016). These genotypes could be elite donors for height and stem diameter to facility mechanical harvesting for elite farmers; meanwhile, genotypes with low height such as L-2955-13 and FA-90 could be used to develop cultivar to traditional farmers where cotton is harvested by hand. These results agreed with Anwar et al. (2002), Copur (2006) and Ashokkumar and Ravikesavan, (2011) who observed significant differences among cultivars for plant height across environments. Similar results of variation were reported among genotypes evaluated under different environments for LY and other agronomic traits, where all the main effects and interaction were significant (Killi and Gencer, 1995; Unay et al., 2004; Killi and Harem, 2006). Genotype-Environment interactions were significant for all traits evaluated, resulting in rank differences of the genotypes. In 1999, a survey of private and public U.S. cotton breeders considered yield stability as the second most important criteria for selecting parents to use in hybridization, only being preceded by yield potential (Bowman, 2000). Analysis of variance using AMMI model indicates the relative magnitude of genetic, environment and their interaction for lint yield in the genotypes evaluated, which allows identification of promising genotypes with different adaption levels. The results showed high statistical significant ($P \leq 0.01$) for genotypes and genotype x environment, providing 23.47 and 65.47% of the total sum of squares, respectively. The GGE biplot for LY of the 16 cotton varieties are shown in Fig. 1. The biplot explained 66.22% of the total variation relative to genotype and GEI, with the first two interaction principal component axes. This figure helps to understand the relationship among environments. Considering the vector length, Ospino during 2013 (OSP13) had the longest vector and ARA14 had the relative short vector. The rest of environments had similar vector lengths. In this study, the angles between the same location but different years, excluding OSP13 and OSP14, were less than 90°, indicating high correlations among them. According to Yan (2002) and Yan and Kang (2003), the angle between vectors of environments indicates the correlation coefficient between them, and their length is a measure to discriminate genotype among environments. Therefore, OSP13 was the most discriminating environment. Ospino and its surroundings represent the target environments for the breeding program for being considered the area with greatest potential for cotton cultivation. Another important part of the genotype evaluation process is selecting the appropriate field trial locations that best represent the target environments for which the breeding program is directed toward (Campbell and Jones, 2005). C-2955-25 was the genotype with the highest yield but is unstable. The genotypes DeltaPine-16, Aleppo-90, FA-90 and Aleppo-118 showed high stability and high yields. On the other hand, BSR-293, Deir Azzor-22 and C-5529-26 showed stability but yield under the average. L-2955-13 was the genotype with higher stability but poor yields (Fig. 2) Previously, Gonzalez et al. (2007) reported similar results, where the PC1 represented 69.0 % of the total sum of squares, and the genotype most stable among 12 environments of Venezuela was FA-90. In this study, it was possible to identify genotypes with high yield and good performance in the other agronomic traits, these genotypes could be incorporated in future breeding program as potential parents to generate new cotton population or inbred lines. Dewdar (2013) indicated that cotton breeders should consider environmental conditions and stability as a criterion for selecting high yielding cultivars.

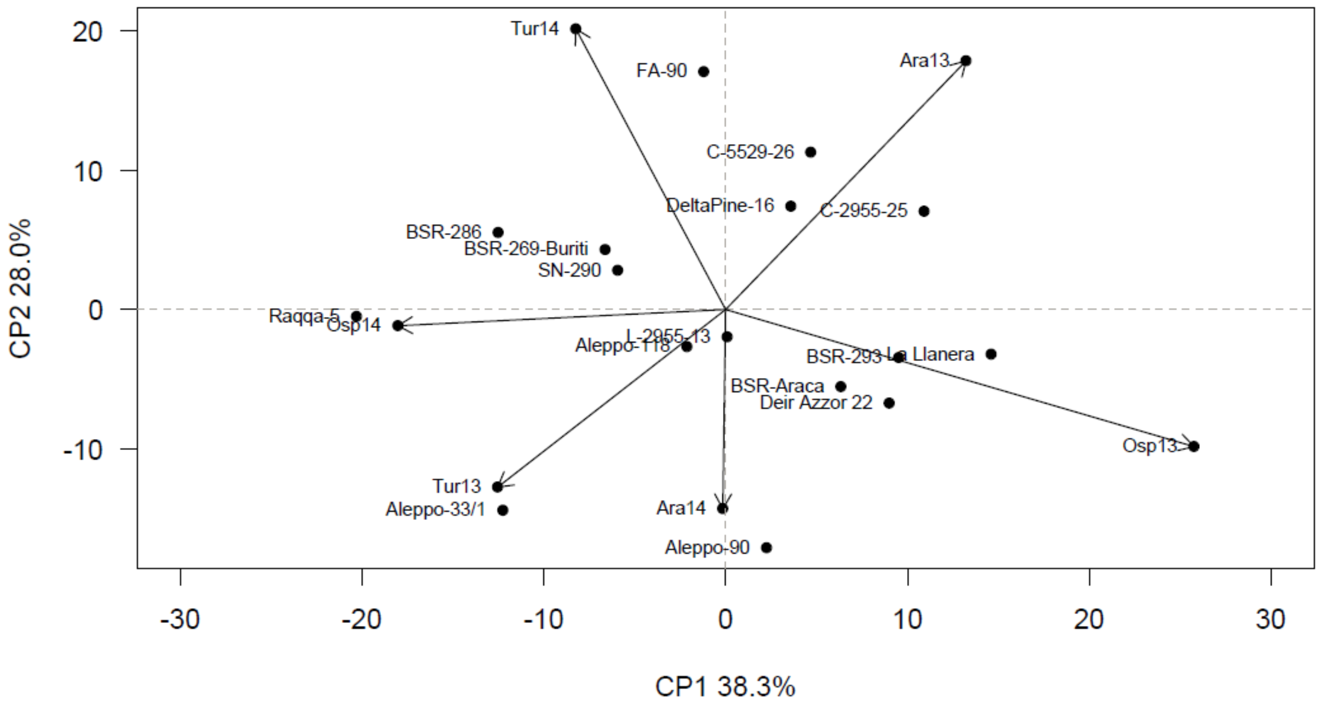

Figure 1. AMMI biplot display PC scores of sixteen cotton genotypes across six environments based on lint yield average

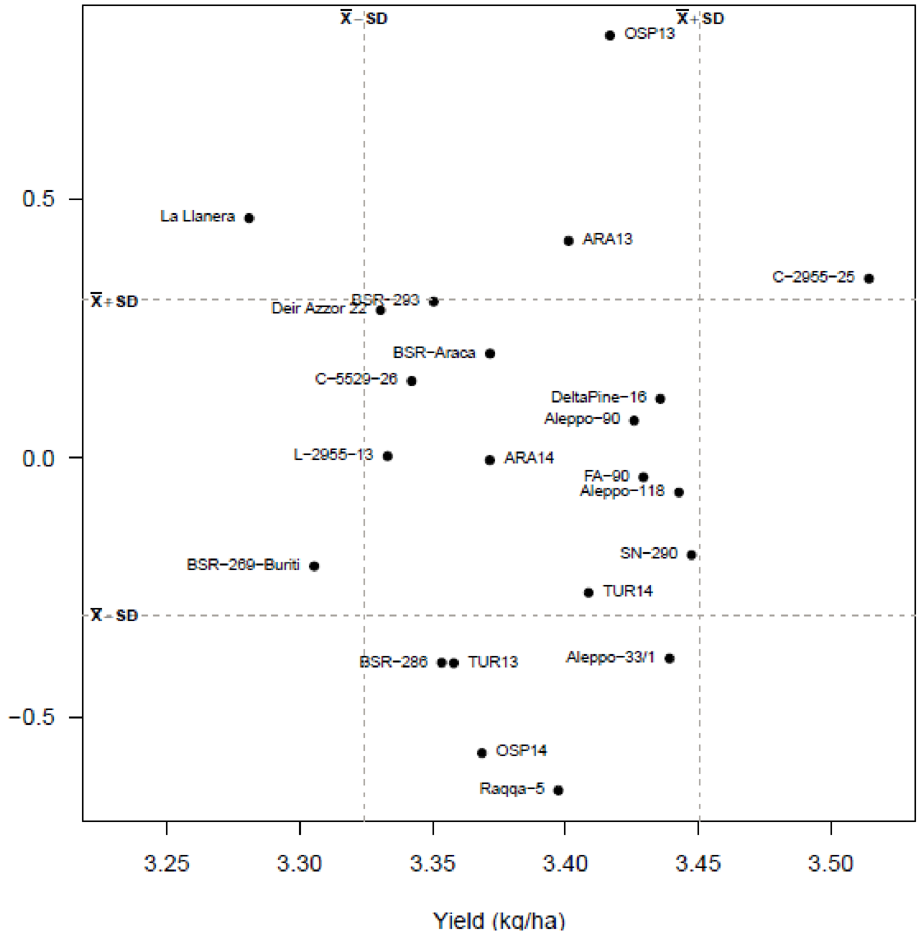

Figure 2. AMMI plot of principal effects and GEI for lint yield of sixteen cotton genotypes across six environments

CONCLUSIONS

Several genotypes used in this study seem to contain alleles that can be used to improve lint yield and other agronomic traits of INIA upland cotton germplasm. The germplasm from Syria and Brazil present a valuable resource to develop varieties with better yield and traits. The best foreign genotypes identified were Aleppo-118, Aleppo-90, from Syria with high lint yield and other agronomic traits of interest. Stability analysis indicated that DeltaPine-16, Aleppo-90, FA-90 and Aleppo-118 were stable and with superior lint yield. L-2955-13 was the most stable genotype but with low LY (3333 kg.ha^{-1}) across environments. The selected genotypes were identified as potential source to incorporate in breeding programs and develop new genotypes in the future, targeting tropical lowlands agroecologies.

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