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Analysis of Genetic Diversity of *Sorghum bicolor* ssp. *bicolor* (L.) Moench using ISSR Markers

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Abstract: Ethiopia is one of the origins and centers of genetic diversity of sorghum. As sorghum ranks fifth in global cereal production, the study of its genetic diversity from its center of origin is important for its improvement. Therefore, the aim of the present study was to evaluate the within and among population genetic diversity of sorghum from Ethiopia using Inter Simple Sequence Repeat markers. Eight individuals from each of eleven sorghum populations of highland, intermediate and lowland ecological zones and one population of improved variety and stay-green cultivars were used for the study. DNA was extracted from young leaves and PCR was conducted using seven primers. A total of 55 clear and reproducible bands with 100% polymorphism were generated. The total genetic diversity (GD) and Shannon's diversity index (I) were 0.205 and 0.296, respectively. North Gondar Intermediate altitude population showed the highest gene diversity (0.3) while the lowest diversity (0.1) was exhibited by North Shewa population. AMOVA showed that 44.45, 30.84 and 24.71% of the total variation was attributed to within populations, among populations and among groups, respectively. Group based UPGMA exhibited four distinct clusters whereas the population based UPGMA showed all populations clustered within its own ecological zone except North Gondar (I_1) intermediate altitude population and improved varieties and stay-green (IM) individuals that were clustered in the highland populations. Intermediate altitude populations exhibited higher genetic diversity than low and high altitude populations indicating these high and low altitude populations may have originated from the intermediates through ecological adaptations.

Key words: Diversity, ecological zones, molecular markers, sorghum

INTRODUCTION

Ethiopia is one of the origins and centers of diversity of Sorghum (*Sorghum bicolor* (L.) Moench). This crop is an important food crop and is widely grown in the highland, lowland and semiarid regions of the country (Abdi *et al*., 2002; Zheng *et al*., 2011). It is important for food, feed, fiber and fuel across a range of agro-ecosystems. Sorghum also used as raw material by industries to produce different products including starch, fiber, dextrose syrup, biofuels and alcohol (Iqbal *et al*., 2010). It is the fifth most important grain crop within the past decade in the world with yearly production of 60 million tones. Among the 44 million hectares of land devoted to global sorghum production, about 90% of it contributed by developing countries with largest share from Africa and Asia (Leder, 2004).

Sorghum is a tight clustering and overlapping C_4 drought tolerant plant with high water use efficiency. Ethiopian sorghum is well known for its high lysine content and grain quality, shoot fly resistance, grain mould resistance and cold tolerance as the result of high genetic diversity. However, this high diversity has been eroded due to adoption of local landraces and imported varieties (Engels and Hawkes, 1991).

Many studies such as, genetic diversity, conservation and breeding were done on several crops using Inter Simple Sequence Repeat (ISSR) marker techniques. This marker is suitable for plant genetic diversity study with high polymorphism and more reproducibility than RAPD marker.

Studies have been done on genetic diversity of sorghum by using different types of markers such as RAPD (Ayana and Bekele, 2000; Prakash *et al*., 2006) and microsatellite (Dje *et al*., 2000). Ejeta *et al*. (2000) used molecular markers in breeding of sorghum. However, there are no studies that were done on genetic diversity of sorghum by using ISSR markers except a single work by

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Fang *et al.* (2008) who studied relations and genetic distance. All of these studies were done using sample pooling method and did not analyze the diversity at individual level. As sorghum is self-pollinating plant, genetic diversity study of each individual within populations from different agroecological zones of its center of origin is important.

Conservation and improvement of this economically valuable crop would have significant effects for sustainable agriculture and food security. The objective of this study was to analyse genetic diversity for conservation and genetic improvement of this plant.

MATERIALS AND METHODS

Plant material: Eleven sorghum populations that were collected from different agroecological zones of Ethiopia were obtained from Institute of Biodiversity Conservation (IBC). Four of those populations were collected from lowland (<1600 m.a.s.l), three from intermediate (1600-1990 m.a.s.l) and four from highland (>1900 m.a.s.l) altitudes (Fig. 1). In addition, one population contains improved varieties obtained from Melkasa Agricultural Research Center and stay-green cultivars from Addis Ababa University, totally representing 12 populations. Each of the 12 populations was represented by eight individuals making the total number of individuals 96. That is, eight individuals from each of the 11 populations and eight additional individuals from the IM population (five improved varieties and three stay-green cultivars). The individual samples were sown in the glasshouse to obtain young leaves for DNA extraction (Table 1).

DNA extraction: Young leaves of about 300 mg were ground in liquid nitrogen and total genomic DNA was isolated by using CTAB method of Wang *et al.* (1996) with minor modifications. Quality of the DNA was checked by electrophoresis of the samples on 1% agarose gel and staining with ethidium bromide. DNA concentration was determined by nanodrop.

PCR amplification and electrophoresis: Twenty primers (Sigma-Aldrich) were screened on individuals of different populations and altitudes and seven primers that produced clear and polymorphic band pattern were selected for further study (Table 2). PCR was carried out in a total volume of 25 μL reaction mixture. The reaction mixture consisted of 2.0 μL of 20 ng DNA, 2.5 μL of 10x PCR buffer A (Himedia, India), 2.5 μL of 25 mM $MgCl_2$, 0.33 μL of 1U firepol Taq DNA polymerase (Solisbiodyne, Estonia), 0.20 μL of 100 mM of each dNTP (Himedia, India), 0.4 μL primer (20 pmol μL^{-1}). Amplification was performed using Techne, Model FTC41H2D Thermal Cycler under optimized temperature condition profiles. Those are initial denaturation step of 4 min at 94°C, followed by denaturation for 30 sec at 94°C, annealing at 45/49°C (depending on the type of primer) for

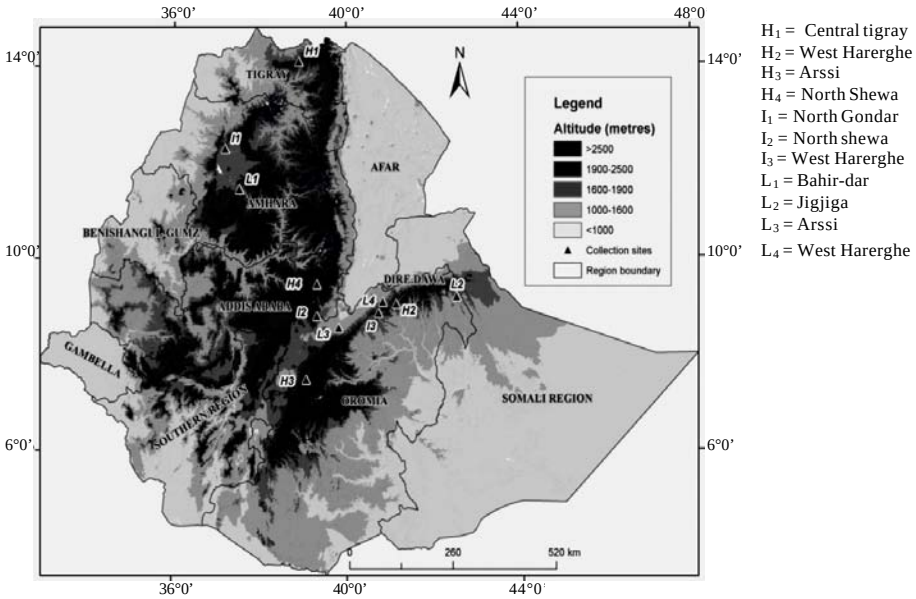

Fig. 1: Map of Ethiopia showing the sampled populations of *Sorghum bicolor* ssp. *bicolor*

Table 1: Description of *Sorghum bicolor* ssp. *bicolor* samples collected from different sites in Ethiopia and improved varieties and stay-green cultivars

Group name	Code	Accession (variety name)	Region	Zone (Release center)	Altitude	Desirable Traits
Highland	H_1	219974	Tigray	Adwa	1920	Normal Cultivar
	H_2	230871	Oromiya	West Harerghe	2020	
	H_3	237029	Oromiya	Arssi	2390	
	H_4	237045	Amhara	North Shewa	2150	
Intermediate						
	I_1	226078	Amhara	North Gondar	1855	
	I_2	237043	Amhara	North Shewa	1570	
	I_3	241200	Oromiya	West Harerghe	1620	
Lowland						
	L_1	226088	Amhara	Bahir-dar	1580	
	L_2	231191	Somali	Jigjiga	1350	
	L_3	231229	Oromiya	Arssi	540	
	L_4	241186	Oromiya	West Harerghe	1570	
IM	IM_1	Sorcoll 141/07	Amhara	North Wollo	1700	High nitrogen and green
	IM_2	Sorcoll163/07	Amhara	North Wollo	1700	leaf area
	IM_4	Sorcoll146/07	Amhara	North Wollo	-	
	IM_3	Melkam	-	Melkassa	-	High yield -Quality seed
	IM_5	B-35	-	ICRISAT		-Drought tolerant
	IM_6	76T#23	-	Melkassa		-Early maturing
	IM_7	Teshale	-	Melkassa/Sirinka	-	
	IM_8	Gambella		Melkassa	-	

H, I, L for Highland, Intermediate and Lowland sorghum populations, IM_1, IM_2 and IM_4 for Stay-green sorghum varieties and IM_3, IM_5, IM_6, IM_7, IM_8 for Improved sorghum varieties

Table 2: Primers used in ISSR analysis, number of loci scored, number of polymorphic loci and size range of the bands

Primer code	Sequence 5'-3'	Total no. of loci	No. of polymorphic loci	Size range (bp)
810	GAGAGAGAGAGAGAGAT	9	9	200-1000
824	TCTCTCTCTCTCTCTCG	8	8	300-1000
825	ACACACACACACACACT	8	8	200-900
827	ACACACACACACACACG	8	8	200-900
841	GAGAGAGAGAGAGAGAYC	9	9	200-1000
873	GACAGACAGACAGACA	7	7	300-900
880	GGAGAGGAGAGGAGA	6	6	300-800
Total		55	55	

Single-letter abbreviations for mixed base positions: Y = (C, T)

45 sec and then extension at 72°C for 2 min. The last cycle had performed with a final extension step of 7 min at 72°C.

Data scoring and analysis: Each ISSR band was considered as an independent locus and polymorphic bands were scored as present (1) or absent (0) for all the 96 individual samples. Only clearly reproducible bands were scored and differences in band intensity were not considered. Data analysis was conducted using only the polymorphic bands. Analysis of Molecular Variance (AMOVA) was carried out in three ways. One was done using the populations grouped in to four groups (highland, intermediate, lowland and IM varieties of cultivated sorghum), the second was done for the 12 populations i.e., using the four groups of cultivated sorghum into 12 populations) the third was using entire populations without grouping. A pair-wise genetic similarity matrix was generated using Jaccard similarity coefficient. A principal coordinate analysis was performed based on Jaccard (1908) for all individuals and a plot was generated using 2D and 3D coordinates.Similarities among the 12 populations were quantified with the Jaccard similarity coefficient and visualized using a cluster analysis, Unweighted Pair-Group Method with Arithmetic averages (UPGMA) and illustrated in a phenogram.

RESULTS

Banding patterns of the ISSR primers: The total number of scored bands varied from six for primer 880 to nine for primers 810 and 841 (Table 2). The three primers 824, 825 and 827 showed the same, eight, number of polymorphic bands and the remaining primer 873 showed seven polymorphic bands with a mean of 8 bands per primer and the size of the bands ranged from 200 to 1000 bp. Figure 2 shows the amplification pattern of primer 824.

All of the four groups of populations (highland, intermediate, lowland and IM), Number of Polymorphic Loci (NPL), Percentage of Polymorphism (PP), Number of Scored Bands (NSB), Genetic Diversity (GD), Standard Deviation (SD) and Shannon Index (I) are shown in Table 3. All the primers showed 100% polymorphism. Among all the 12 populations, I_1 population from North Gondar exhibited the highest PP (76.36%) followed

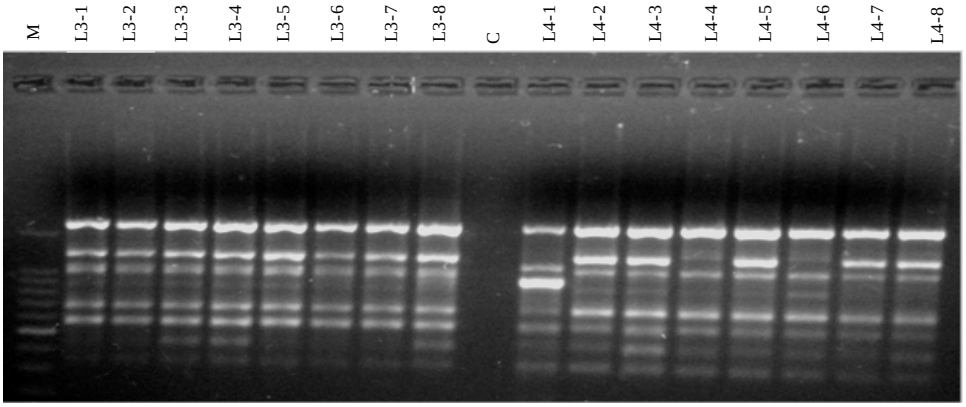

Fig. 2: ISSR band patterns of *Sorghum bicolor* ssp. *bicolor* from Arssi (L_3) and West Harerghe (L_4) populations using primer 824. M: DNA ladder, C: Control, $L_{3\text{-}1}$ to $L_{3\text{-}8}$: Individuals for L_3 population, $L_{4\text{-}1}$ to $L_{4\text{-}8}$: Individuals for L_4 population, L_3 & L_4: Populations collected from Arssi and Hararghe, respectively

Table 3: Number of scorable bands (NSB), Number of polymorphic loci (NPL), Percentpolymorphism (PP), Shannon index (I), Gene diversity (GD) of the 12 populations of sorghum using the seven primers

Primers	NSB	NPL	PP	GD±SD	I±SD
810	9	9	100	0.40±0.10	0.575±0.116
824	8	8	100	0.37±0.12	0.552±0.144
825	8	8	100	0.38±0.12	0.559±0.134
827	8	8	100	0.46±0.06	0.654±0.061
841	9	9	100	0.42±0.10	0.605±0.121
873	7	7	100	0.37±0.56	0.558±0.100
880	6	6	100	0.42±0.08	0.610±0.084
Total	55	55	100	0.40±0.10	0.600±0.110

NSB: Number of scorable bands, NPL: Number of polymorphic loci, PP: Percent polymorphism, GD: Genetic diversity, I: Shannon Index and SD: Standard Deviation

Table 4: Measures of genetic diversity in the 12 populations of sorghum

Populations	NPL	PP	GD±SD	I±SD
219974 (H_1)	23	41.82	0.170±0.210	0.244±0.301
230871 (H_2)	33	60.00	0.240±0.220	0.350±0.300
237029 (H_3)	29	52.73	0.210±0.230	0.310±0.310
237045 (H_4)	14	25.45	0.100±0.200	0.145±0.260
226078 (I_1)	42	76.36	0.300±0.200	0.420±0.270
237043 (I_2)	25	45.45	0.200±0.210	0.262±0.300
241200 (I_3)	22	40.00	0.151±0.210	0.220±0.300
226088 (L_1)	29	52.73	0.201±0.210	0.300±0.303
231191 (L_2)	30	54.55	0.210±0.202	0.305±0.300
231229 (L_3)	30	54.55	0.200±0.204	0.300±0.300
241186 (L_4)	35	63.64	0.260±0.200	0.380±0.300
IM*	35	63.64	0.214±0.210	0.320±0.300
Total			0.205±0.210	0.296±0.295

*Improved and stay-green, H_1: Central Tigray sorghum population, H_2: West Harerghe, H_3: Arssi, H_4: North Shewa I_1: North Gondar, I_2: North Shewa, I_3: West Harerghe, L_1: Bahir-dar, L_2 Jigjiga, L_3 Arssi, L_4 West Harerghe, NPL: Number of polymorphic loci, PP: Percent polymorphism, GD: Genetic diversity, I: Shannon Index and SD: Standard Deviation

by IM (63.64%) and L_4 (63.64%) from West Hararghe while the least percentage of polymorphism was found in H_4 population (25.45%) from North Shewa.

I_1 of Gondar showed highest genetic diversity of 0.3 whereas L_4 and H_2 of west Hararghe populations exhibited 0.26 and 0.24, respectively. The lowest genetic diversity was observed in North Shewa (H_4) population with GD of 0.1 (Table 4).

Analysis of molecular variance (AMOVA): Analysis of molecular variance revealed that highest percentage of variation is attributed to variation within populations with 44.45% and variation among the 12 populations with 30.84%, respectively. The least variation is among the four groups (highland, intermediate, lowland and IM) groups with 24.71%. Generally, the results of AMOVA revealed patterns of genetic diversity and supports the larger

Table 5: Analysis of Molecular Variance (AMOVA) among groups, among populations and within populations of cultivated sorghum in Ethiopia

Source of variation	d.f.	Sum of squares	Variance components	Variation (%)
Among groups	3	130.615	1.26375	24.71
Among populations	8	119.146	1.57743	30.84
Within populations	84	191.000	2.27381	44.45
Total	95	440.760	5.11499	100

d.f: degree of freedom

Table 6: Pairwise Jaccard coefficient similarity matrix of the four groups of sorghum (IM, intermediate, lowland and highland groups)

	Highland	Intermediate	Lowland	IM
Highland	-			
Intermediate	0.407	-		
Lowland	0.628	0.516	-	
IM	0.236	0.380	0.224	-

IM: Improved and stay-green sorghum varieties

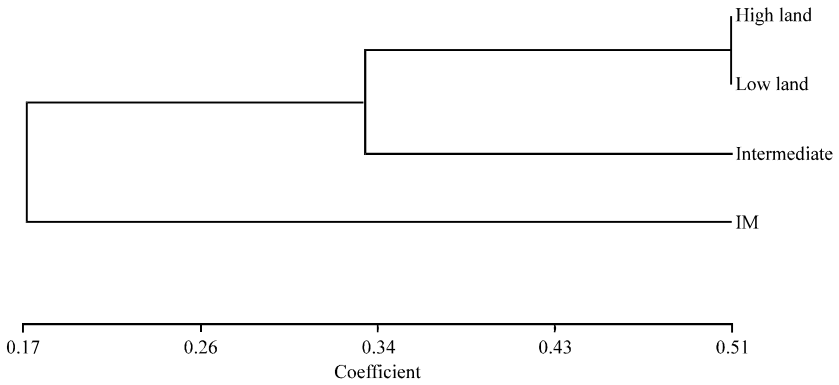

Fig. 3: ISSR-based UPGMA dendrogram for populations of sorghum based on four ecological zones. IM = improved+stay-green varieties of sorghum

genetic diversity found within the populations rather than among populations and among groups is the least (Table 5).

Cluster analysis: UPGMA analysis based ecological zones of the four groups of sorghum varieties revealed two major groups in the dendrogram (Fig. 3). The first cluster contains the highland, lowland and intermediate groups and two sub clusters within the first major cluster, i.e., highland and lowland clustering together which was confirmed by highest Pairwise Jaccard's similarity of 0.628 (Table 6). However, the intermediate group formed its own separate cluster in the second sub-cluster. Most of the populations collected from the same ecological zones tend to form their own grouping except the intermediate population of Gondar (I_1) intermixing with highland and IM groups (Fig. 4). Population based UPGMA Pairwise Jaccard's similarity also confirmed this in which the populations with similar ecological zones exhibited high similarity matrix. L_2 and L_3 populations showed the highest similarity matrix of 1.0 (Table 7).

Similarly, UPGMA analysis of individual collected from similar ecological zones tended to form their own grouping. Some intermediate I_1 individuals intermixing with highland, putative stay greens and the lowland individuals. Moreover, similar patterns of grouping in the case of NJ were also observed but with more intermixing than UPGMA clustering (Fig. 5). Surprisingly, the individuals of lowland populations that were collected from different localities clustered together. However, in the case of NJ analysis, some intermediate individuals from north Shewa (I_2) and west Hararghe (I_3) populations clustered with lowland populations whereas few individuals from the three remaining groups (highland, intermediate and IM) intermixed with each other.

Principal coordinate analysis: The first three coordinates of the PCO having eigenvalues of 7.30, 4.85 and 3.74 with variance of 9.76%, 6.5 and 5.014, respectively were used to show the grouping of individuals (Fig. 6). The three groups of populations, Highland, intermediate and lowland, formed separate clusters. However, the four lowland populations (L1, L2, L3 and L4) and some intermediate and highland individuals were intermixed throughout the populations.

Table 7: Pairwise Jaccard similarity coefficient-based comparisons among 12 populations of sorghum

	H-1	H-2	H-3	H-4	I-1	I-2	I-3	L-1	L-2	L-3	L-4	IM
H-1	-											
H-2	0.543	-										
H-3	0.538	0.557	-									
H-4	0.591	0.512	0.610	-								
I-1	0.433	0.573	0.514	0.536	-							
I-2	0.378	0.311	0.332	0.344	0.328	-						
I-3	0.281	0.240	0.250	0.251	0.246	0.502	-					
L-1	0.507	0.435	0.489	0.531	0.440	0.463	0.344					
L-2	0.476	0.461	0.432	0.480	0.432	0.474	0.381	0.563	-			
L-3	0.476	0.461	0.432	0.480	0.432	0.474	0.381	0.563	1.000	-		
L-4	0.481	0.393	0.419	0.434	0.377	0.393	0.335	0.553	0.589	0.589	-	
IM	0.469	0.443	0.449	0.529	0.519	0.272	0.227	0.396	0.39	0.390	0.353	-

IM: Improved and stay-green sorghum, H_1: Central Tigray sorghum population, H_2: West Harerghe, H_3: Arssi, H_4: North Shewa I_1: North Gondar, I_2: North Shewa, I_3: West Harerghe, L_1: Bahir-dar, L_2: Jigjiga, L_3: Arssi, L_4: West Harerghe

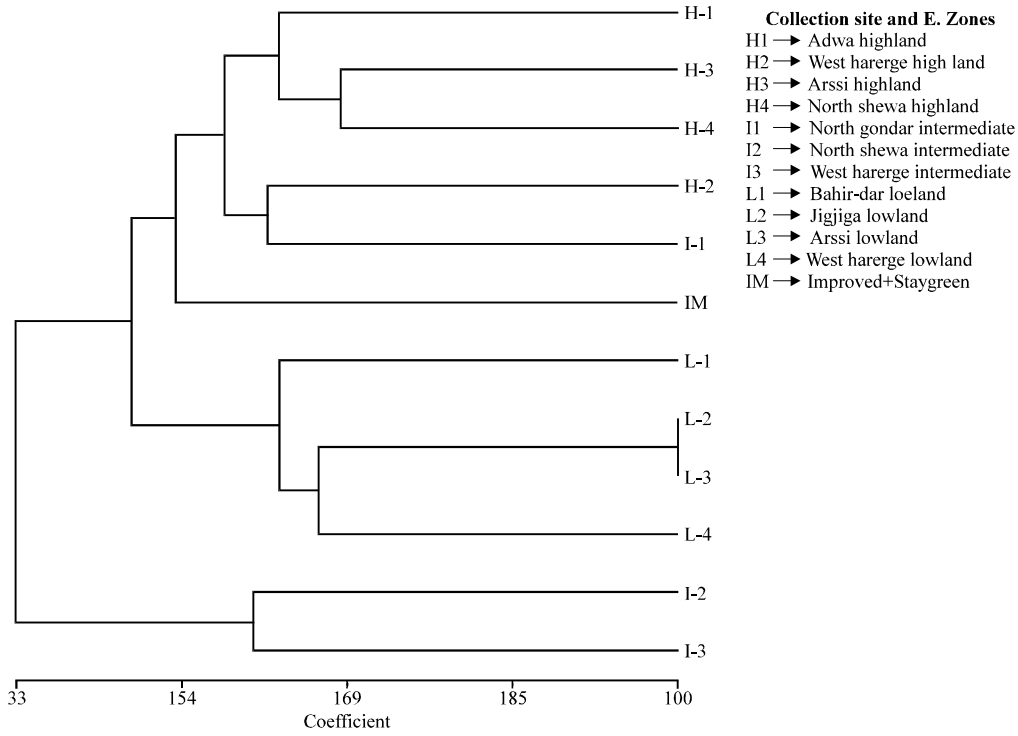

Fig. 4: ISSR-based UPGMA dendrogram for 12 populations of sorghum

DISCUSSION

In the present study, all the diversity parameters confirm that there is higher genetic diversity within populations than among populations and among groups of sorghum. Furthermore, I_1 sorghum population from intermediate altitude of North Gondar with genetic diversity of 0.30 and Shannon index of 0.42 found to be the most diverse population. On the other hand, H4 highland population of North Shewa exhibited the lowest genetic diversity with genetic diversity of 0.10 and 0.15 Shannon index. This is in agreement with the reports of Ohsawa and Ide (2008) on comparative analysis of genetic variation in plant species along vertical and horizontal gradients on mountains. According to these authors, high altitude populations have less diversity than lower and intermediate altitude populations whereas the intermediate altitude populations have greater diversity than populations at low and high altitudes. This is probably because of intermediate altitude contains geographically central populations under optimal environmental conditions but the peripheral populations are in suboptimal situations.

The result of this study is also in line with the reports of Mekbib *et al.* (2009) that altitude has an impact on genetic diversity in Ethiopia and shapes on-farm genetic

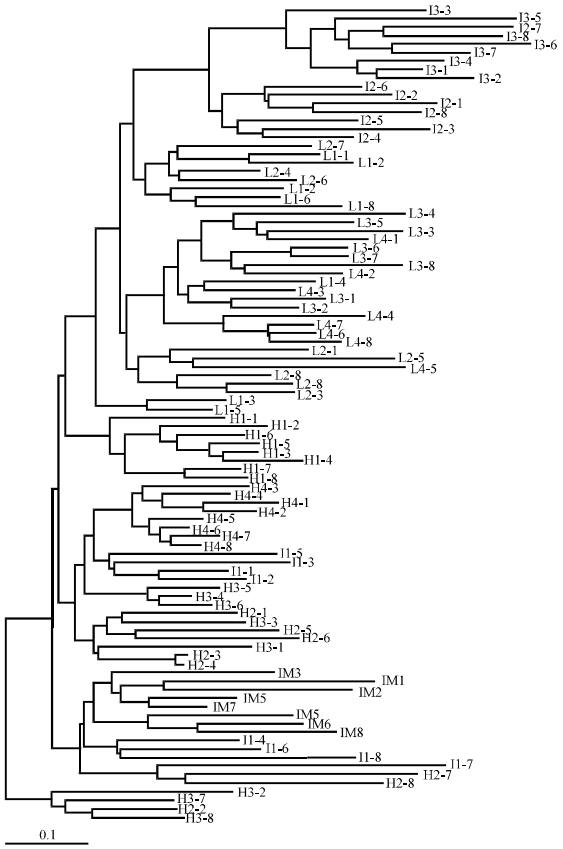

Fig. 5: Neighbor-joining analysis of 96 individuals based on Jaccard's coefficient

resources of sorghum. The lowest sorghum diversity occurred in the highland and lowland; where the highest was in the intermediate, i.e., it assumed that sorghum moved from the intermediate to both highlands and lowlands through ecotype differentiation. Availability of optimum temperature and water could be another reason for the higher genetic diversity in the intermediate altitudes. The temperature is relatively low on the highlands and water is scarce in the lowlands. Additionally, the reason for the low diversity in the lowlands is because of the genetic base of lowland sorghum is narrow in Ethiopia (Mekbib *et al*., 2009). There may be great drop in genetic diversity for genes involved in domestication because of sever bottlenecks during domestication and founding effect (small initial population) of crop cultivars. The IM (63.64%), lowland (L_4 = 63.64%) and highland (H_2 = 60%) of West Harerghe populations exhibited higher Percent Polymorphism (PP) and Shannon's index (I) next to North Gondar (I_1) population as compared to other sorghum populations. Genetic diversity of sorghum could be influence by human factors including markets and government policies related to land ownership and natural factors such as altitude, soil, climate and gene flow with wild sorghum (Price *et al*., 2005; Teshome *et al.,* 2007; Tesso *et al*., 2008). According to Kudadjie (2006), geneflow could arise from hybridization between different seed lots of the same variety or between different varieties.

AMOVA of this result showed that there is higher genetic variation within populations (44.45%) than among populations within groups (30.84%) and among groups (24.71%). Nguni *et al.* (2011) reported a highly significant genetic variation both within and among accessions of sorghum from Zambia using SSR markers. However, contrary to the results of the present study by using ISSR markers, among population diversity was higher than within population diversity. Although, the UPGMA showed a clear grouping and differentiation based on populations of ecological zones and locality, in the neighbor-joining tree, some individuals were intermixed all over the populations. Therefore, this high genetic diversity within population is expected in individual-based UPGMA and neighbor-joining tree just like the

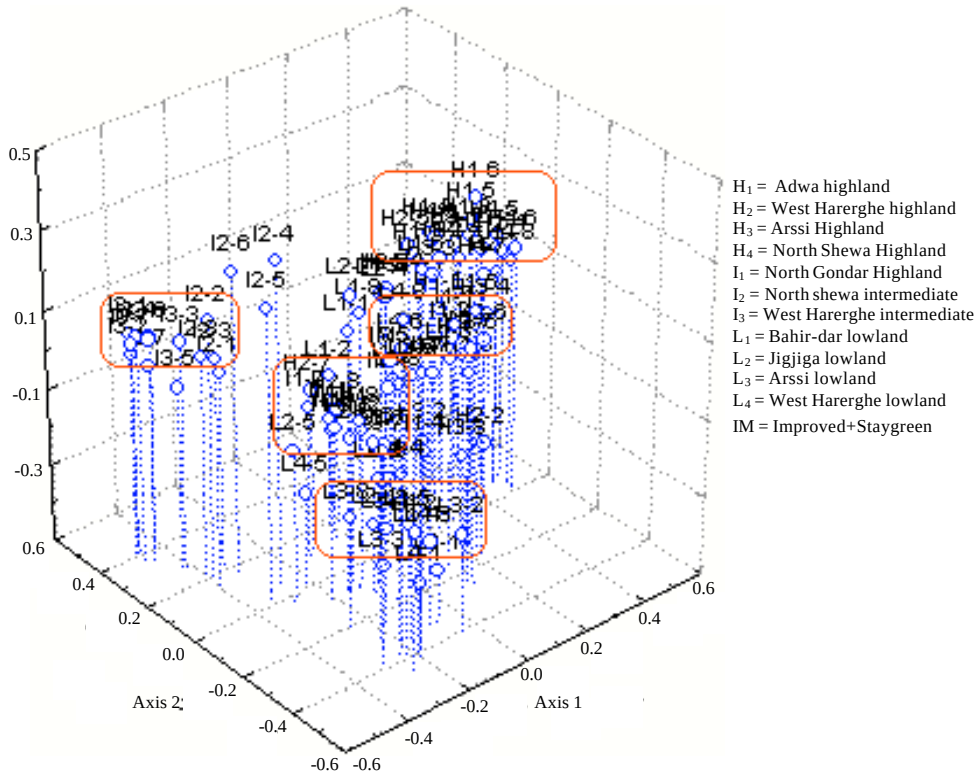

Fig. 6: Three-dimensional representation of principal coordinate analysis of 96 individual samples of 12 populations of sorghum investigated with ISSR

highest genetic diversity within populations rather than among populations and among groups of the AMOVA result.

The reason for this high genetic diversity within population could be due to high seed exchange across horizontal or vertical localities and ecological zones in the form of human activities such as cropping and markets, which could lead to intermix within the populations. Unlike other cereals, sorghum is not restricted to a given locality or ecological zone rather it is widely exchanged among local communities and markets as well as is cultivated across all altitudes. In a similar way, Tsehaye *et al.* (2009) pointed out that when crops fail due to drought, a poor family particularly women migrate temporarily to distant places to glean the stocks and panicles that fall to the ground during harvesting season. This phenomenon encourages the gene flow among populations and individuals of sorghum and is a common practice in south Tigray and usually called qarmia.

The UPGMA analysis showed that each group formed a discrete cluster based on populations from similar ecological zones. There are two sub clusters within the first major cluster i.e., highland and lowland clustering together in group-based UPGMA. This clustering may formed because of intermediate sorghum varieties adaptation to both highland and lowlands through ecotype differentiation and developed cold and drought tolerant for highland and lowland conditions, respectively just as reported by Mekbib *et al.* (2009). Similarly, populations that were collected from similar ecological zones tended to form their own grouping except the North Gondar (I_1) population that was intermixed with highland and IM groups. Gene flow through seed movement by human activities across similar ecological zones could be the most possible reason for this UPGMA clustering. For example, different localities of lowland sorghum populations were clustered together. Jigjiga population (L_2) frequently experiencing severe drought, showed a significant level of genetic similarity with geographically non-adjoining L_3 Arssi populations. This indicates that these populations adapt lowland environmental condition rather than highland or intermediate condition irrespective of the geographic distance. The other probable reason for these impressive genetic similarities within the lowland populations could relate with massive seed movements. This movement is associated with response to recurrent drought donation by relief agencies such as NGOs and government organizations across the low land dry areas.

North Gondar (I_1) intermediate individuals intermixed with highland (H_4, H_3 and H_2 individuals of north Shewa, Arssi and west Hararghe, respectively). Those I_1 North Gondar individuals also intermixed with putative stay-green individuals (IM_1 and IM_4 of north Wello individuals). I_1 intermediate altitude (PP = 76.36%) and IM (PP = 63.64%) populations that have highest percent polymorphism intermixed with the highland populations. This could be adaptation for the cold highland environmental condition with optimum rainfall rather than the dry and water deficient lowlands. As a result, the IM populations (improved and putative stay-greens) and I_1 populations clustered with the highland populations that have high biomass and higher grain yield. It is also obvious that the IM and intermediate individuals have a potential to adapt and tolerate the harsh environmental conditions of lowlands.

In agreement with McGuire (2000), formal breeding, starting with the Ethiopian Sorghum Improvement Program in the early 1970s, has mainly sought with open-pollinated modern varieties. For example, 'Seredo', Tanzanian variety released to Ethiopia in the 1980s, suitable for highland environment of good rainfall availability with long maturity, gives high biomass and grain yields. The Ethiopian Seed Enterprise does not multiply lowland sorghum seed because of low yield. Therefore, research stations are the only formal suppliers of small amount of modern seed varieties to farmers in lowland areas through regional agricultural offices and non-governmental organizations. As the program developed, it expanded from exclusive focus on highlands of the country and now structures work along the three broad ecological zones (low, mid and high-altitude). Moving landraces within and/or across similar ecological zones could be a powerful way of improving production stability. Seed exchange network could establish as a means of facilitating access to locally adapted sorghum genetic resources (Tsehaye *et al*., 2009).

PCO analysis showed similar result to UPGMA except the individuals of lowland group populations intermixed throughout the IM and intermediate populations. The reason for this intermixing could be lowland sorghum individuals may easily adapt to the intermediate altitude (1600-1900 m.a.s.l) that the lowland populations tolerate the water deficit condition so that they perform better if they get intermediate ecological zone with better conducive environment. PCO based on individuals of IM (improved and putative stay-green) population was not found to form its own separate cluster. The reason for this intermixing could be the improved varieties produced from different genetic background parents of those ecological zones.

CONCLUSION

The present study showed that intermediate altitude populations of cultivated sorghum exhibited higher genetic diversity than populations at low and high altitudes. These highland and lowland sorghum populations may have originated from the intermediates through ecological adaptation. All the diversity parameters confirmed that there is high genetic diversity within populations than among populations. Regarding the four groups of cultivated sorghum populations based on altitudes, the genetic diversity pattern exhibited the pattern of higher diversity within populations than among populations and among populations diversity higher than among groups diversity.

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