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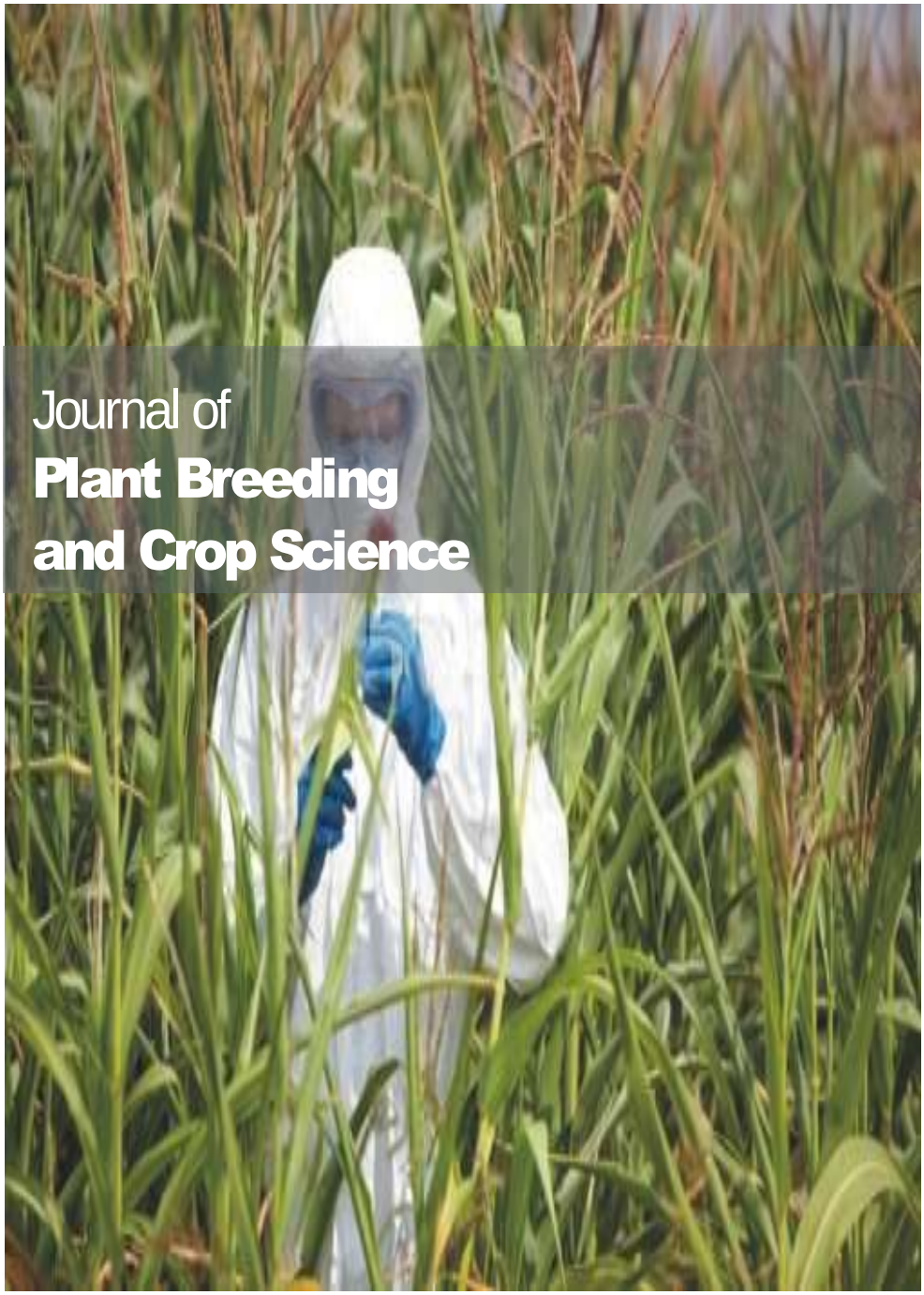

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Journal of Plant Breeding and Crop Science

Full Length Research Paper

Assessment of salt tolerance and variability within some rice germplasm using microsatelittes

Anyomi W. E.*, Ashalley R., Amoah N.K.A, Blay E.T. and Ofori K.

Department of Crop Science, College of Basic and Applied Sciences, University of Ghana, P. O. Box Lg 44, Legon, Accra, Ghana.

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Soil salinity is a major abiotic stress that affects rice production. It can reduce yield drastically and result in total crop failure. The objectives of this study are to determine the genetic diversity within thirty-six rice accessions and to identify genotypes that are tolerant to salinity. Thirty-six rice accessions including three check varieties were sown in experimental pots and their leaves harvested for DNA extraction. Screening was done with 31 simple sequence repeats (SSRs) primers, of which 14 were markers for salt tolerance, 2 primers did not produce any results. 28 out of the 31 primers were polymorphic. The polymerase chain reaction (PCR) products were run and visualized on a 3% agarose gel matrix stained with ethidium bromide. Amplified bands were scored and analyzed with PowerMarker v3.25 and DARwin v5 software. The genetic diversity among the accessions assembled was high (He=0.6, I=0.516, PIC=0.471). Saltol primers RM10711 and RM10793 were the only primers able to completely discriminate tolerant genotypes from susceptible ones, hence they can be used in selections involving the genotypes. Accessions SR1, IR72, Sebota 337-1, Perfume (Short) type, Anyofula, Local Red, GR18Red, GH1580, GH1528, GH1575, NericaL23, NericaL24 and NericaL27 performed well under salinity stress in this study and were identified to be superior among the accessions used. These accessions should be incorporated into major breeding programs to improve the salt tolerance of existing commercial lines or for the production of new commercial lines.

Key words: Rice, abiotic stress, agarose gel, genetic diversity, polymerase chain reaction (PCR), simple sequence repeats (SSRs) markers, salinity.

INTRODUCTION

Rice (*Oryza sativa* L. and *O. glaberima* Steudl) is an important cereal of the Poaceae family grown worldwide. Two species of importance in the genus are namely, *O. sativa* the universally cultivated Asian rice, and *Oryza glaberrima,* the West African cultivated rice. African rice is now only rarely grown in pure stands. It is currently grown in mixture with the Asian rice in various proportions. The extent of even this form of mixed cultivation is diminishing as it is being replaced with 'pure' Asian rice (Nayar, 2010). Rice is the fastest growing food source in Africa (Nwanze et al., 2006). Rice has become a major staple crop in recent decades with a per capita consumption of 25 kg/annum in Ghana, but most of the consumption is met by imports (MOFA, 2010). In 2009,

*Corresponding author. E-mail: lordedems@gmail.com.

Ghana imported over 350,000 tons of milled rice worth 600 million US dollars (Duffuor, 2009).

Salinity of arable land is one of the most important factors retarding rice growth and development at both vegetative and reproductive stages (Zeng and Shannon, 2000; Zeng et al., 2003). Salinity is expected to have devastating global effects, resulting in 30% land loss within the next 25 years, rising to 50% by the year 2050 (Wang et al., 2003). Salinity reduces plants ability to take up water and results in growth reduction. Excess salts in plants can reach toxic levels, which causes premature leaf senescence and ultimately photosynthesis reduction (Munns, 2002). Specific effects of salt stress on plant metabolism, especially on leaf senescence, have been related to the accumulation of toxic Na+ and Cl- ions and to K+ and Ca^{2+} depletion (Al-Karaki, 2000). Salinity associated with excess NaCl adversely affects the growth and yield of plants by depressing the uptake of water and minerals and normal metabolism (Akhtar et al., 2001; Akram et al., 2001).

Salinity has been found to negatively impact a number of yield components of rice including stand establishment; panicles, tillers and spikelets per plant; floret sterility; individual grain size; and even delayed heading. Maas and Grattan (1999) and Hanson et al. (1999) indicated that rice yields decrease by 12% for every unit (dsm^{-1}) increase in EC (average root-zone EC of saturated soil extract) above 3.0 dsm^{-1}. Plant breeding harnesses inherent variability within plants for economic gains. It involves redistributions of genes in a population. In so doing, genes of interest are propagated whiles others are eliminated from the population.

In breeding for salinity tolerant varieties there is a need to screen and select tolerant varieties. The success of salt tolerance breeding programs employing traditional screening and selection has some limitations. Conventional methods of plant selection for salt tolerance are difficult because of the large effects of the environment and low narrow sense heritability of salt tolerance (Gregorio, 1997). Genetic improvement of salt tolerance in rice using marker assisted selection (MAS) is most feasible and promising strategy (Munns, 2002). SSR are ideal genetic markers (Farooq and Azam, 2002) and have a repeat-unit length of 1-6 base pair units arranged in repeats of mono-, di-, tri-, tetra and penta-nucleotides (A,T, AT, GA, AGG, AAAG) with different lengths of repeat motifs. According to Mason (2015) and Joshi et al. (2011), SSRs are highly informative, codominant, multi-allele genetic markers that are experimentally reproducible and transferable among related species. The variation in the number of tandemly repeated units results in highly polymorphic banding pattern (Farooq and Azam, 2002) which are detected by PCR, using locus specific flanking region primers where they are known. Microsatellite markers have been used to identify the variation among rice cultivars (Yang et al., 1994; Akagi et al., 1997; Garland et al., 1999). Similarly, Thanh et al. (1999) showed the genetic variation identified by microsatellite markers to be useful in evaluating upland rice accessions. Genetic diversity between parental genotypes is usually estimated by measurements of physiological and morphological differences of quantitative and economically important traits. Diversity ensures a large genepool from which traits can be mined for economic gains. Without diversity, a species finds it difficult to adapt to the ever changing environmental and biotic stresses. With wide range of crops, a breeder is able to screen and choose materials for various purposes. The objective of this study was therefore to determine diversity among thirty-six accessions of rice and to identify salt tolerant genotypes using SSR markers.

MATERIALS AND METHODS

Experimental site

The research was carried out at the Biotechnology Center of the Department of Crop Science, University of Ghana, Legon.

Experimental materials

Plant materials were obtained from the Plant Genetic Resources Research (PGRRI) Institute and Savanna Agricultural Research Institute (SARI), both for the Council for Scientific and Industrial Research (CSIR), Ghana. NERICA rice genotypes were also obtained from Africa Rice Center, Sahel Station- Senegal, and from farmers' fields in Ghana. Table 1 shows the list of accessions and their sources. FL478 an International salt-tolerant accession and IR29, an international sensitive genotype obtained from Africa Rice Center were used as checks for salinity. Accession 'CG14' from Africa Rice Center was included as check for O. *glaberrima*, in the accessions from African countries. A total of 36 germplasm was investigated (Table 1).

Sowing of the rice accessions

The seeds of the 36 accessions of rice were nursed in a nursery pot at the green house in the department of crop science, Legon. The leaves of the accessions were collected for DNA extraction.

DNA extraction for molecular studies

DNA was extracted using E.Z.N.A. ™ SP Plant DNA Mini Kit. Approximately 0.03 g of leaf samples was frozen in liquid nitrogen and ground in a microfuge tube. 400 µl of buffer SP1 was immediately added followed by 5 µl of RNase. The samples were incubated at 65°C for 10 min. 140 µl of buffer SP2 was added to each sample and mixed vigorously by vortexing. This was followed by incubation on ice for 5 min and centrifugation at 14000 rpm for 10 min. The supernatant that resulted was carefully aspirated into an Omega® Homogenizer Column placed in 2 ml collection tube and centrifuged at 14000 rpm for 2 min. 500 µl of the clear lysate that resulted was transferred into a 1.5 ml tube. Binding conditions of the sample were then adjusted by pipetting 750 µl of buffer sp3/ ethanol mixture directly onto the clear lysate.

650 µl of the resulting mixture was transferred into a Hiband® DNA Mini Column placed in a 2 mL collection tube and centrifuged

Table 1. The germplasm studied and their sources.

Accession number	Name	Source
1	GH 1593	CSIR-PGRI, Ghana
2	GH 1575	CSIR-PGRI, Ghana
3	GH1585	CSIR-PGRI, Ghana
4	GH1598	CSIR-PGRI, Ghana
5	GH1571	CSIR-PGRI, Ghana
6	GH 1533	CSIR-PGRI, Ghana
7	GH 1528	CSIR-PGRI, Ghana
8	GH1545	CSIR-PGRI, Ghana
9	GH 1580	CSIR-PGRI, Ghana
10	GH 1599	CSIR-PGRI, Ghana
11	SR-1	ARC- Senegal
12	IR-29	ARC-Senegal (Susceptible check)
13	CG14	ARC- Senegal (*O. glaberrima*)
14	FL478	ARC- Senegal (Tolerant check)
15	Nerica L23	ARC- Senegal
16	Nerica L9	ARC- Senegal
17	Nerica L24	ARC- Senegal
18	Nerica L27	ARC- Senegal
19	Sebota 33	Cameroun
20	Sebota 337-1	Cameroun
21	Perfume (Short type)	Thailand
22	Sebota 41	Cameroun
23	Local Red	Farmer collection, E/R
24	Anyofula	CSIR-PGRI, Ghana
25	Good and New (JP)	Japan
26	IR 72 (Ph)	IRRI, Philippines
27	GH 1837	CSIR-PGRI, Ghana
28	Matigey	CSIR-PGRI, Ghana
29	Basmati 122	IRRI, Philippines
30	GR 18 Red	CSIR-SARI, Ghana
31	Local Basmati-2	IRRI, Philippines
32	Koshihikari	Japan
33	Viwornor	CSIR-PGRI, Ghana
34	Sebota 281-2	Cameroun
35	Sebota 68	Cameroun
36	Abidjan	Local farmer

for 1 min at 14000 rpm. Later, the flow through was discarded. This was repeated for the remaining mixture. The columns were then placed into a new 2 ml collection tube and 650 µl of SPW Wash Buffer diluted with ethanol was added. This was centrifuged at 14000 rpm for 1 min and the flow through was discarded. This step was repeated with the sample volume of SPW wash buffer. The empty column was centrifuged at 14000 rpm for 2 min. The Hiband® Mini column was then transferred into a sterile 1.5 ml tube and 100 µl of pre-warmed (65°C) elution buffer was added. This was then centrifuged at 14000 rpm for 1 min to elute the DNA.

Polymerase chain reaction (PCR) amplification

Thirty-One SSR primers (Table 2), 15 of which were markers for salt tolerance were selected for the PCR. 15 µl PCR reaction was composed of 1X Taq buffer, 2 mM $MgCl_2$, 1U Taq DNA polymerase, 0.2 mM of each dNTP's and 0.4 µM SSR primer pair. Deionized water was used to make up the volume to the final PCR reaction volume. The thermal cycling conditions were as follows; 94°C for 5 min followed by 35 cycles of 94°C for 1 min, 1 min for annealing temperature (55-67°C) depending on the primer used and 72°C for 2 min, and a final extension at 72°C for 5 min. Primers were selected from previous works of Deepti et al. (2013), Huyen et al. (2012) and Thompson et al. (2010).

Gel electrophoresis of amplified products

For a 120 ml electrophoresis casting tray, 3.6 g of agarose was weighed into 120 ml of TAE buffer. The initial weight was noted. This was then melted on a hot plate after which distilled water was used to make up the weight difference. The melted gel was then cooled under running water after which 8 µl of ethidium bromide

Table 2. SSR primers used in PCR, their annealing temperatures and flanking sequences.

Primer	Ann. temp (OC)	Forward sequence (5'- 3')	Reverse SEQ. (5'-3')
RM20	55	ATCTTGTCCCTGCAGGTCAT	GAAACAGAGGCACATTTCATTG
RM307	55	GTACTACCGACCTACCGTTCAC	CTGCTATGCATGAACTGCTC
RM5	55	TGCAACTTCTAGCTGCTCGA	GCATCCGATCTTGATGGG
RM552	55	CGCAGTTGTGGATTTCAGTG	TGCTCAACGTTTGACTGTCC
RM19	55	CAAAAACAGAGCAGATGAC	CTCAAGATGGACGCCAAGA
RM454	55	CTCAAGCTTAGCTGCTGCTG	GTGATCAGTGCACCATAGCG
RM11	55	TCTCCTCTTCCCCCGATC	ATAGCGGGCGAGGCTTAG
RM518	55	CTCTTCACTCACTCACCATGG	ATCCATCTGGAGCAAGCAAC
RM334	55	GTTCAGTGTTCAGTGCCACC	GACTTTGATCTTTGGTGGACG
RM237	55	CAAATCCCGACTGCTGTCC	TGGGAAGAGAGCACTACAGC
RM259	55	TGGAGTTTGAGAGGAGGG	CTTGTTGCATGGTGCCATGT
RM474	55	AAGATGTACGGGTGGCATTC	TATGAGCTGGTGAGCAATGG
RM178	67	TCGCGTGAAAGATAAGCGGCGC	GATCACCGTTCCCTCCGCCTGC
RM489	55	ACTTGAGACGATCGGACACC	TCACCCATGGATGTTGTCAG
RM312	55	GTATGCATATTTGATAAGAG	AAGTCACCGAGTTTACCTTC
RM253	55	TCCTTCAAGAGTGCAAAACC	GCATTGTCATGTCGAAGCC
RM336	55	CTTACAGAGAAACGGCATCG	GCTGGTTTGTTTCAGGTTCG
RM10655	55	AGTACCGTTGAATCCGATATGC	TGGTTGAGGTGCTGAATTGG
RM10696	60	CCTTCGACTCCATGAAACAAACG	TCTCTTTGCCCTAACCCTATGTCC
RM10711	55	GCTTCGATCGATGAGAAAGTAGAGG	GAATCTCCCATCCTTCCCTTCC
RM10713	60	ATGAACCCGGCGAACTGAAAGG	CTGGCTCCCTCAAGGTGATTGC
RM10748	60	CATCGGTGACCACCTTCTCC	CCTGTCATCTATCTCCCTCAAGC
RM10722	60	GCACACCATGCAAATCAATGC	CAGAAACCTCATCTCCACCTTCC
RM10793	60	GACTTGCCAACTCCTTCAATTCG	TCGTCGAGTAGCTTCCCTCTCTACC
RM10800	60	CGTACGCCCTCACATCACCTTTCC	CTCTCCGGGAGCTCACTTGTCG
RM10825	60	GGACACAAGTCCATGATCCTATCC	GTTTCCTTTCCATCCTTGTTGC
RM10843	60	CACCTCTTCTGCCTCCTATCATGC	GTTTCTTCGCGAAATCGTGTGG
RM10852	60	GAATTTCTAGGCCATGAGAGC	AACGGAGGGAGTATATGTTAGCC
RM10864	60	GAGGTGAGTGAGACTTGACAGTGC	GCTCATCATCCAACCACAGTCC
RM10890	60	GCTTCGGCTCTTCATTCACTGG	GCGATTATAGGAGCGCTATGTGG
RM10927	60	TGGATCCCACTAATCCAAATGC	GAAAGACTCCTTCCAATGTTAGGC

was added and swirled gently to mix. The agarose gel was then poured into the casting tray and combs set in place. This was allowed to solidify for 40 min. The casting tray together with the solidified agarose was then transferred into the electrophoretic tank and submerged with TAE buffer and the combs gently removed. 10 µl of PCR amplicons was mixed with 3 µl of 6X loading dye and carefully loaded into wells created by removing the combs. The leads of the electrophoretic tank were connected to electrophorese amplicons at 100V for 1 h after which the gel was viewed with the aid of a UV transilluminator.

Allele scoring and data analysis

The polymorphic bands were scored for each of the microsatellite primer pairs in each genotype based on presence 1 or absence 0 for bands to generate a matrix of 1 and 0. The size (in nucleotide base pair) of the amplified band for each SSR marker was determined based on its migration with comparison to a known molecular weight marker (1Kb DNA Ladder). Allele numbers, gene diversity, heterozygosity and polymorphic information content (PIC) were calculated with Power Marker v3.25 software (Liu and Muse 2005). Between samples, genetic distances were assessed through simple matching index as implemented in DARwin v5 software (Perrier et al., 2003). A dendogram was constructed based on the unweighted pair-group method with arithmetic averages (UPGMA) using the neighbor-joining (NJ) method as implemented in the same software. Polymorphic information content (PIC) values were calculated with the following formula (Anderson et al., 1993):

$$PIC_i = 1 - \sum_{j=1}^{n} P_{ij}^2$$

Where, n is the number of marker alleles for marker *i* and P*ij* is the frequency of the *j*th allele for marker *i.*

RESULTS

Microsatellite variations of the rice accessions

Salt tolerance SSRs

Twenty saltol SSR markers were screened; 16 produced

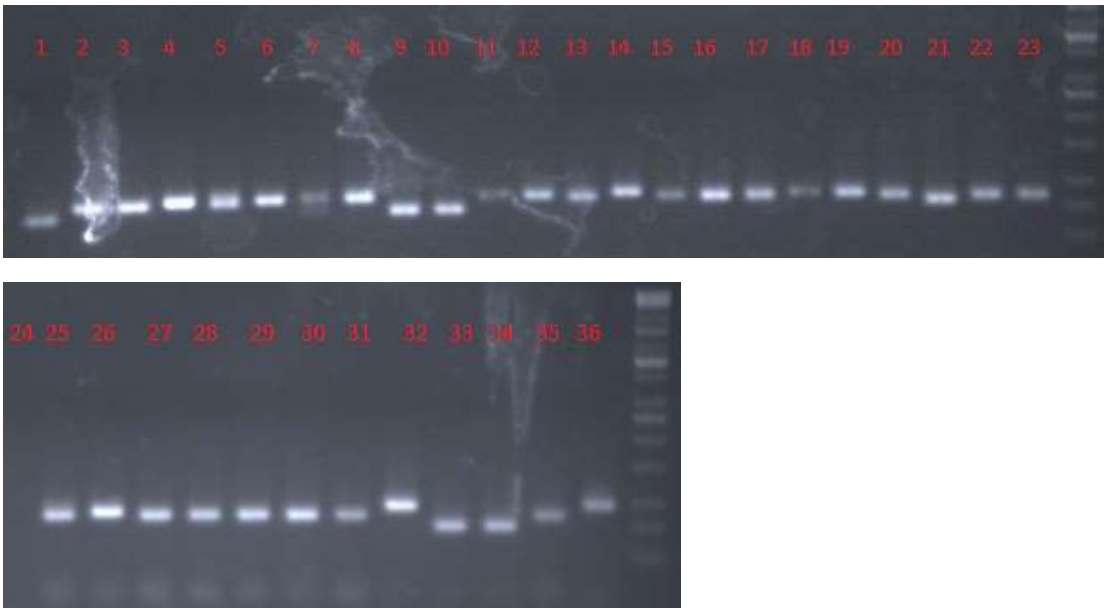

Plate 1. A gel image of the banding pattern of the genotypes with Primer RM10711. 1= Local Red, 2= Sebota 41, 3= Perfume (Short type), 4= Sebota 337-1, 5= Sebota 33, 6= Nerica L27, 7= Nerica L24, 8= Nerica L9, 9= Nerica L23, 10= FL478, 11= CG14, 12= IR-29, 13= SR-1, 14= GH1599, 15= GH1580, 16= GH1545, 17= GH1528, 18=GH1533, 19= GH1571, 20= GH1598, 21= GH1585, 22= GH1575, 23= GH1593, 24= Abidjan, 25= Sebota 68, 26= Sebota 281-2, 27= Viwornor, 28= Koshihikari, 29= Local Basmati-2, 30= GR 18 Red, 31= Basmati 122, 32= Matigey, 33= GH 1837, 34= IR 72 (Ph), 35= Good and New (JP), 36= Anyofula.

polymorphic bands. Primers RM10711 and RM10793 showed bands that differentiated the tolerant and susceptible checks. Primer RM10711 (Plate 1) was able to separate ten Ghanaian, three Nericas, two Basmati and three Sebota entries as susceptible. It also distinguished Koshihikari as susceptible. The primer recognized three entries as tolerant. Accessions Perfume (Short type), Sebota 41, Good and New (JP), GH 1528, Sebota 337-1, local Red, GH1545, GH 1580, SR-1 and Gh1585 did not share any bands with the two checks.

Primer RM10793 showed that accessions GH1599, SR-1, GH1571, GH1533, Nerica L9, Nerica L27, Perfume (Short type), GH1837, Matigey, Basmati 122, Sebota 281-2, Sebota 68 shared a common band with IR29 the susceptible check. GH 1575, GH1585, GH1598, GH 1528, GH1545, GH 1580, CG14, Nerica L23, Nerica L24, Sebota 33, Sebota 41, Local Red, Good and New (JP), IR 72 (Ph), Local Basmati-2, Koshihikari, Viwornor all had similar bands to FL478, the salt tolerant check. GH1593, Sebota 337-1, Anyofula, and GR 18 Red did not share bands with the two checks in relation to primer RM10793 (Plate 2).

Diversity among accessions

Out of the 31 SSR primers used, 28 produced polymorphic bands representing 84.8%. Fourteen out of the 28 SSR primers were located at the saltol loci of the rice genome, the remaining 14 spread throughout the entire genome. A total of 116 alleles with an average of 4.14 alleles per locus were generated by the 28 primers (Table 3).

The highest allele frequency was 100% produced by primer RM454 and the lowest allele frequency was 30% produced by primers RM10748, RM10864 and RM20. The overall average allele frequency was 60%. The Polymorphic Information Content, PIC, of the primers among the 36 rice genotypes was observed in the range of 0.053 to 0.785, with an average of 0.471. The genetic diversity within the population was 51.6% but RM20 had the highest diversity of 84.6% and RM454 the lowest of 5.4%. Primer RM20 had the highest PIC of 0.829 followed by RM10864 and RM10793 respectively, with RM454 having the least PIC of 0.053. Primer RM20 had the highest diversity discrimination of 84.6% followed by RM10864 with 78.5% and RM10793 with 75% respectively; while RM454 had the lowest diversity of 5.4%.

Genetic divergence of rice population as revealed by the dendrogram

A dendrogram was constructed based on the unweighted pair-group method with arithmetic averages (UPGMA) using the neighbor-joining (NJ) method. The dendrogram constructed grouped the accessions into clusters,

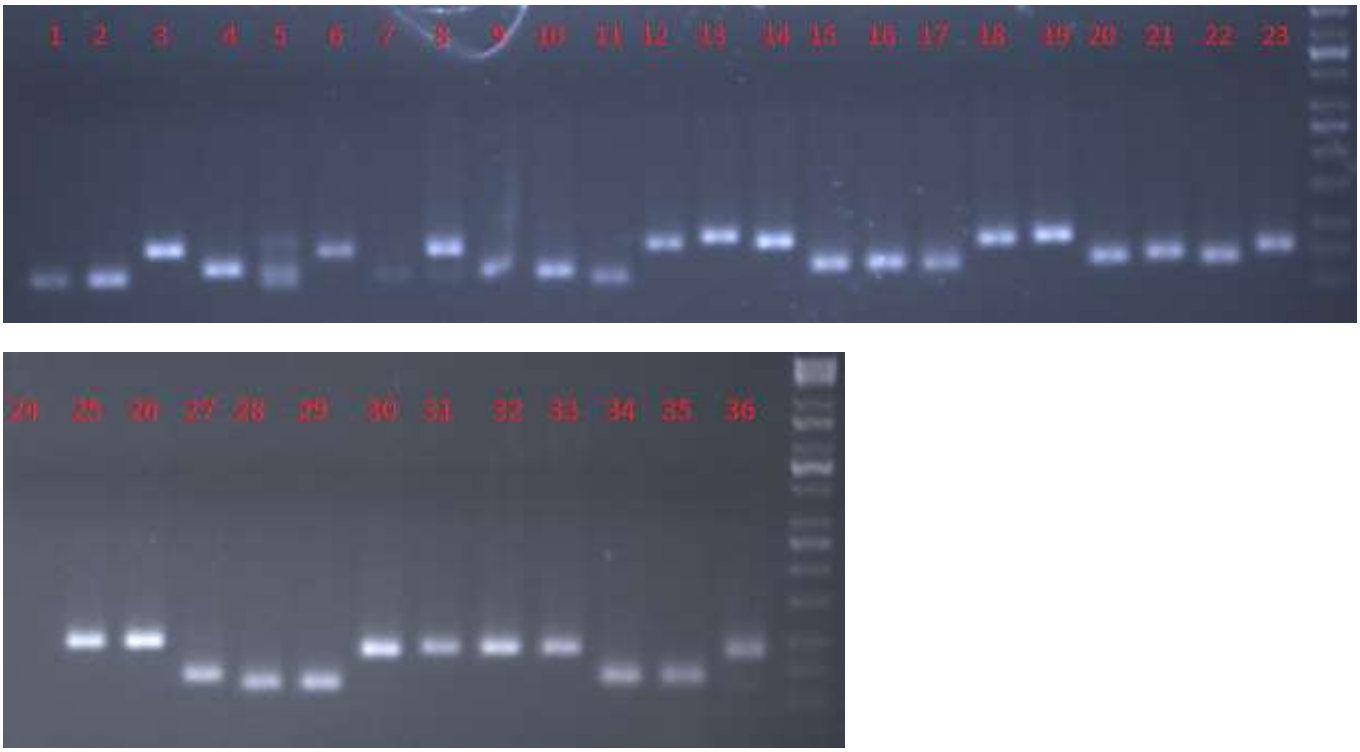

Plate 2. A gel image of the banding pattern of the genotypes with primer RM10793. 1= Local Red, 2= Sebota 41, 3= Perfume (Short type), 4= Sebota 337-1, 5= Sebota 33, 6= Nerica L27, 7= Nerica L24, 8= Nerica L9, 9= Nerica L23, 10= FL478, 11= CG14, 12= IR-29, 13= SR-1, 14= GH1599, 15= GH1580, 16= GH1545, 17= GH1528, 18=GH1533, 19= GH1571, 20= GH1598, 21= GH1585, 22= GH1575, 23= GH1593, 24= Abidjan, 25= Sebota 68, 26= Sebota 281-2, 27= Viwornor, 28= Koshihikari, 29= Local Basmati-2, 30= GR 18 Red, 31= Basmati 122, 32= Matigey, 33= GH 1837, 34= IR 72 (Ph), 35= Good and New (JP), 36= Anyofula.

indicating the diversity that existed between the accessions. The accessions were separated into three major clusters (Figure 1). Major Cluster 1 comprised Nine Ghanaian entries, all four Nerica's, one Sebota entry, one Thailand entry, one Japanese entry and the two checks. Major cluster 2 comprised eight Ghanaian entries including a collection from a farmer's field, four Sebota entries, the two Basmati entries from the Philippines, one Japanese entry and two entries from ARC Senegal. Major cluster 3 had only IR72 (Ph) from the Philippines. Major cluster 1 had two sub clusters. Sub cluster I and II, sub cluster I had two sub clusters, the first contained three Nerica entries and five Ghanaian entries together with the checks, the second cluster consisted of four Ghanaian entries only. All Nericas were separated from each other in this sub group. Sub cluster II had only two entries, Nerica L24 and Local Red

Major cluster 2 had two sub clusters III and IV. Sub cluster III had two lower clusters; the first comprised six Ghanaian entries, one Japanese and Philippine entry. The second contained only one entry from Sebota. Sub cluster IV had two sub groups; the first consisted of three Sebota entries, two Ghanaian entries and one Philippine entry. The second group had one entry from ARC Senegal.

DISCUSSION

Genetic diversity and identification of most informative markers

Out of the 31 SSR primers used, 28 produced polymorphic bands representing 90.03%. Fourteen out of the 28 SSR primers were located at the saltol loci of the rice genome, the remaining 14 were spread throughout the entire genome. A total of 116 alleles with an average of 4.14 alleles per locus were generated by the 28 primers. The average allele frequency was 0.6. The PIC ranged from 0.053 to 0.829, with an average of 0.471. The genetic diversity within the population was 51.6%. The highest diversity was 84.6% and the lowest was 5.4%. Similar results were recorded by Lang et al. (2008) where 95% of SSR markers for genetic diversity were reported to be polymorphic in IR64 variety. The results are also in line with Deepti et al. (2013), who reported a higher average PIC of 0.67 for 26 SSR markers within a range of 0.50 to 0.89. The number of alleles obtained per locus was 7.1, which was higher than the values obtained in this work. Mahalingam et al. (2013), on the other hand, reported an average PIC value of 0.44 lower than what was obtained in this study. Their highest PIC value

Table 3. SSR primers used with their parameters for diversity.

Marker	Major allele frequency	No of alleles	Gene Diversity	PIC
RM336	0.4	5	0.690	0.639
RM10655	0.4	5	0.708	0.660
RM10696	0.7	5	0.511	0.470
RM10711	0.4	5	0.719	0.668
RM10713	0.8	4	0.336	0.317
RM10722	0.4	6	0.728	0.689
RM10748	0.3	4	0.742	0.694
RM10793	0.4	7	0.750	0.711
RM10800	0.5	4	0.642	0.583
RM10825	0.5	4	0.640	0.592
RM10843	0.7	4	0.444	0.409
RM10852	0.5	4	0.625	0.568
RM10864	0.3	7	0.785	0.755
RM10890	0.7	2	0.444	0.346
RM10927	0.7	2	0.424	0.334
RM253	0.8	3	0.323	0.285
RM518	0.9	2	0.153	0.141
RM312	0.9	2	0.105	0.099
RM489	0.8	3	0.403	0.363
RM474	0.5	5	0.660	0.604
RM259	0.8	4	0.412	0.383
RM11	0.6	3	0.537	0.441
RM454	1.0	2	0.054	0.053
RM19	0.7	3	0.415	0.349
RM5	0.6	2	0.486	0.368
RM20	0.3	9	0.846	0.829
RM307	0.8	5	0.298	0.287
RM522	0.6	5	0.576	0.536
Mean	**0.6**	**4**	**0.516**	**0.471**

reported was greater than 0.60 and the lowest was 0.035; both lower than what was obtained in this experiment. The present estimate of PIC was also larger than that reported by Hashimoto et al. (2004) in a Japanese rice population comprising 171 cultivars used in brewing of Japanese rice wine; it had a diversity of 0.33.

Singh et al. (2011), in their genetic diversity study of rice genotypes using 30 SSR markers, noted fewer alleles (83) with a lower average of 2.76 alleles per marker; but they had a high PIC value varying from 0.54 to 0.96. Studies by Chakravarthi and Naravaneni (2006) revealed that primer RM20 on chromosome 12 had seven alleles. In the present study, primer RM20 had 9 alleles, indicating that it is very polymorphic and suitable for diversity studies. El-Malky et al. (2007) used 14 microsatellites to generate a total of 122 alleles with an average PIC of 0.782 and a range of 0.438 to 0.891. All their diversity parameters were higher than those obtained in this work. Islam et al. (2012) detected a total of 168 alleles; the number of alleles per locus ranged from 2 to 6 which was lower than what was obtained in this work (2 to 9); but they had an average of 4.2 alleles per locus slightly higher than the value obtained in this study.

Polymorphic information content (PIC) value varied from 0.21 to 0.76 with an average of 0.57 higher than that of this study, 0.471. Lapitan et al. (2007) reported higher parameters than those obtained in this study. They had a total of 176 alleles. Their number of alleles per marker was high ranging from 6 to 22, with an average of 14.6 alleles per locus. Their primers were thus very useful in distinguishing the germplasm used. Roychowdhury et al. (2013) also detected a total of 122 alleles which was higher than that obtained in this study but the primer used had a lower allele range of 2 to 5 alleles compared to this study (2 to 9). They also reported a lower average of 3.21 alleles per locus but the PIC value was 0.524 which was higher than the results from this study.

Emon et al. (2015) reported a total of 209 alleles among 5 rice genotypes using 160 SSR markers. They had a

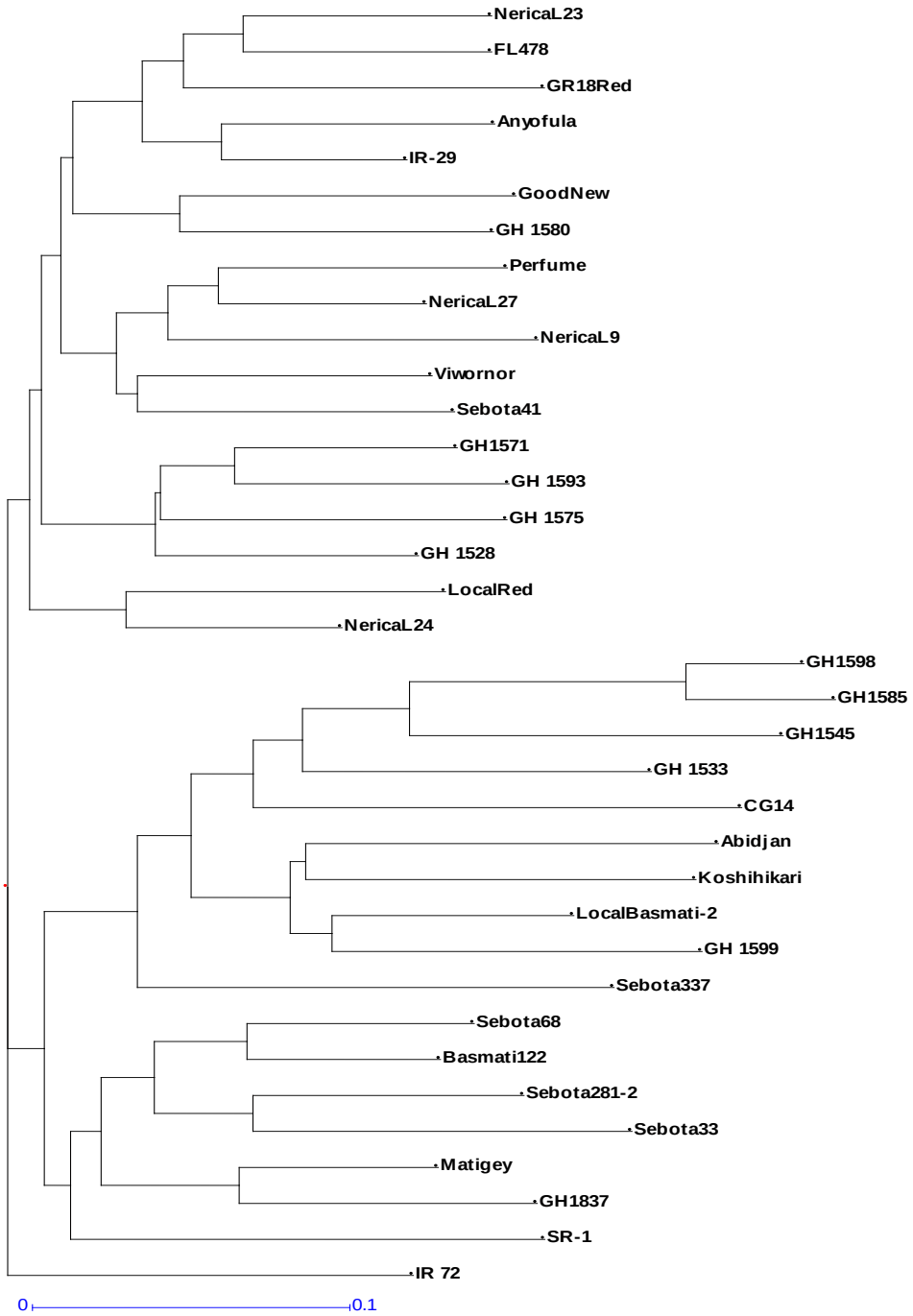

Figure 1. Dendrogram of the clustering of the genotypes with the SSRs markers.

lower PIC of 0.32 and also a lower diversity of 0.37. Ram et al. (2007) reported number of alleles per locus varied from 3 to 8, with average number of alleles per locus at 4.86. This indicates almost the same magnitude of diversity with reference to the markers used in this investigation. Behera et al. (2012) observed a total of 169 alleles, of which 166 were polymorphic from a set of 36 microsatellite markers. Their number of alleles per locus ranged from 2 to 9 with an average of 4.69 alleles per locus. Their PIC ranged between 0.24 and 0.956 with an average of 0.811 per locus, which were all higher than that reported in this work.

High PIC value of a marker indicates high probability to detect the number of alleles among cultivars. A PIC value higher than 0.50 indicates high degree of polymorphism. Based on this RM20, RM10864, RM10793, RM10748

and RM10722 were very good primers for this diversity study. The total number of alleles generated by the 28 primers agrees with findings of Zeng et al. (2004) who observed a total of 123 alleles among 33 rice genotypes with an average of 4.9 alleles per locus. The number of alleles per locus ranged from 2 to 9. The PIC values for the microsatellite loci ranged from 0.06 to 0.85 with an average of 0.57. Low PIC values were observed for 5 primers and the PIC values for the remaining 17 microsatellite loci were all above 0.50. Lapitan et al. (2007) also obtained PIC ranges from 0.18 to 0.91 with an average 0.68 per marker, making them very useful for genotypic studies. Prabakaran et al. (2010) had a total of 11 alleles detected by 5 SSR primers and the number of alleles per locus ranged from 2 to 3 with an average of 2.2 per locus. Among the primers used, RM 481 identified higher number of alleles and average PIC was 0.43. Behera et al. (2012) observed a wider range of PIC, between 0.24 and 0.956 with a higher average of 0.811 per locus than was obtained in this study. The results of this study show that the markers used are revealing and good for genetic diversity studies in rice. Microsatellites are efficient and cost-effective to use. Compared with other markers, they are abundant, co-dominant, highly reproducible and interspersed throughout the genome. In particular, microsatellite markers have been widely applied in rice genetic studies as they are able to detect high levels of allelic diversity. SSR markers are playing important role in identifying genes for salt tolerance that can be helpful for plant breeders to develop new cultivars. Molecular markers could be used to tag QTL and evaluate their contributions to the phenotype by selecting for favorable alleles at those loci in marker assisted selection (MAS) scheme with the aim to accelerate genetic advancement in rice. This is faster, more efficient and cost-efficient than conventional screening under saline field conditions (Gregorio, 1997; Aliyu et al., 2011). The findings in this study imply a great genetic resource for improvement to salinity of rice in Ghana. SSRs discovered here can be incorporated in breeding programs to improve rice materials for farmers.

Selection of salt tolerant genotypes

Progress in rice breeding for salt tolerance entails identifying the major locus with salt tolerance at different growth stages. Out of 14 saltol primers screened, only primers RM10711 and RM10793 were able to discriminate tolerant genotypes from susceptible ones. Based on Primer RM10711, Nerica L23, Local Red, IR 72 (Ph) were tolerant to salinity stress, CG14, Nerica L9, Nerica L24, Nerica L27, Sebota 33, Anyofula, Matigey, Basmati 122, GR 18 Red, Local Basmati-2, Koshihikari, Viwornor, Sebota 281-2, Sebota 68, GH 1593, GH 1575, GH1598, GH1571, GH 1533, GH 1599 were however sensitive to salinity stress. With regard to RM10793, GH1575, GH1585, GH1598, GH1528, GH1545, GH 1580, CG14, Nerica L23, Nerica L24, Sebota 33, Sebota 41, Local Red, Good and New (JP), IR 72 (Ph), Local Basmati-2, Koshihikari, Viwornor were tolerant and GH 1599, SR-1, GH1571, GH 1533, Nerica L9, Nerica L27, Perfume (Short type), GH 1837, Matigey, Basmati 122, Sebota 281-2, Sebota 68 were susceptible accessions. This indicates that the primers could be used in marker assisted selection involving these genotypes. Aliyu et al. (2011) used RM10793 on a collection of 150 diverse rice genotypes with a tolerant salt variety Pokkali and found the marker very informative. Deepti et al. (2013) also found the primer very informative in their study on salt tolerance in some rice accessions. Huyen et al. (2012) used RM10793, RM10711 in Introgressing salinity tolerance QTLs Saltol into AS996 rice variety with five hundred BC2F1 individuals. Kabir et al. (2008) also used twelve SSR markers for parental survey and among them three polymorphic SSR markers, OSR34, RM443 and RM169 were selected to evaluate 26 F3 rice lines for salt tolerance. With respect to marker OSR34, 15 lines were identified as salt tolerant, 9 lines were susceptible and 2 lines were heterozygous. Several SSR primers (RM21, RM51 and RM127) were used by Sohrawardy et al. (2008) for the identification of salt tolerant rice lines of PNR-519 x IR29 in F3 population. Islam et al. (2008) selected different SSR primers to evaluate F2/F3 rice lines for salt tolerance and identified 15 rice lines as salt tolerant by using RM231 and RM24 primers.

Clustering of genotypes

Dendrogram generated by SSR primers further grouped the germplasm into three major clusters. Cluster 1 had IR29 and FL478 clustering together; it also had the Nerica's at different sub clusters. Nerica L9 is a cross between TOG5681 and 3 rounds of backcross to IR64, Nerica L-23 and Nerica L-24 are crosses between TOG5681 and 2 rounds of backcross to IR64 followed by crosses to IR31851-96-2-3-2-1, Nerica L-27 crosses between TOG5681 and 4 rounds of backcross to IR64. This probably explains why they were in the first cluster together, even though at different sub clusters. FL478 is a salt tolerant variety developed from a cross between Pokkali and IR29. The genotypes that clustered closely to these checks were similar to them.

Major cluster 2 had GH1598, GH1585, GH1545, GH 1533, clustering with CG14, a glaberrima and native to Africa, at a lower clustering level. This means the varieties in this group could be indigenous landraces from Africa and may carry a lot of unexploited genes for rice breeding. The glaberrima carries genes for tolerance to a lot of natural stresses, from environmental to biotic stresses (Takeoka, 1965; Second, 1984). Abidjan, Koshihikari, Local Basmati-2 and GH 1599 also formed a cluster; Koshihikari is a known Japanese elite variety,

with cold tolerant genes and the Basmatis are known for their aroma. Sub clustering under this group showed SR-1 singly and separately clustering with the group, Sebota 68, Basmati 122, Sebota 281-2, Sebota 33, Matigey, and GH 1837. SR-1 seed shows shattering tendency when mature. This may imply that the group clustering with it could behave similarly. There is also the possibility that SR-1 is the only one with the shattering ability, hence on a different branch. Shattering is a negative trait in rice breeding. Major cluster 3 had only IR72 which had high amylose content; it is popular among Cambodian farmers because it produces higher yields with superb grains of good quality, long grain and good taste, and can grow in dry season too. This accession did not cluster with any of the accessions indicating how unique and diverse it was from the rest.

The germplasm from PGRRI showed high degree of variability indicating how widely diverse they are genetically and how rich the germplasm is. This is good for rice breeding as it indicates a rich array of genes which could be useful for improving the crop. These results further highlighted the divergence of the population studied. This diversity can be explored in breeding to improve local rice cultivars. Microsatellite markers were able to distinguish between salt tolerant and susceptible entries.

Conclusion

The high PIC value of RM20, RM10864, RM10793, RM10748 and RM10722 and their ability to separate the rice germplasm suggests their usefulness in diversity studies. Genotypes GR18Red, GH1580, GH1528, GH1575, Anyofula, Local Red and NericaL23, NericaL24 and NericaL27 were selected by the markers to be tolerant to salinity stress. They would therefore be candidate genotypes for further development of improved varieties.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Akagi H, Yokozeki Y, Inagaki A (1997). Highly polymorphic microsatellites of rice consist of AT repeats and a classification of closely related cultivars with these microsatellite loci. Theoretical and Applied Genetics 94:61-67.

Akhtar S, Wahid A, Akram M, Rasul E (2001). Effect of NaCl salinity on yield parameters of some sugarcane genotypes. International Journal of Agriculture and Biology 3:507-509.

Akram M, Hussain M, Akhtar S, Rasul E (2001). Impact of NaCl salinity on yield components of some wheat accessions/varieties. International Journal of Agriculture and Biology 4:156-158.

Aliyu A, Adamu AK, Muazu S, Alonge SO (2011). Tagging and Validation of SSR markers to Salinity Tolerance QTLs in Rice (*Oryza* spp). International Conference on Biology, held in Singapore pp. 328-332.

Al-Karaki GN (2000). Growth, water use efficiency and sodium and potassium acquisition by tomato cultivars grown under salt stress. Journal of Plant Nutrition 23:1-8.

Anderson JA, Churchill GA, Autrique JE, Tanksley SD, Sorrels ME (1993). Optimizing parent selection for genetic linkage maps. Genome 36(1):181-186.

Behera L, Patra BC, Sahu RK, Nanda A, Sahu SC, Patnaik A, Rao GJN, Singh ON (2012). Assessment of genetic diversity in medicinal rice's using microsatellite markers. Central Rice Research Institute, Cuttack-753 006, Odisha, India. Australian Journal of Crop Science 6(9):1369-1376.

Chakravarthi BK, Naravaneni R (2006). SSR marker-based DNA fingerprinting and diversity study in rice (*Oryza sativa*. L). African Journal of Biotechnology 5(9):684-688.

Deepti D, Sasidharan N, Macwana S, Sudeshna C, Trivedi R, Ravikiran R, Shah G (2013). Molecular Characterization of Rice (*Oryza Sativa L*) Genotypes for Salt Tolerance Using Microsatellite Markers. The Bioscan 8(2):499-502.

Duffuor K (2009). Budget Statement and Economic Policy of the Government of Ghana for the 2010 Fiscal Year.

El-Malky MM, Fahmi AI, Kotb AA (2007). Detection of genetic variability using microsatellites in rice (*Oryza sativa*. L) African Crop Science Conference Proceedings 8:597-603.

Emon R, Gregorio GB, Adedze N, Islam MM, Islam MR, Ye-Yang F (2015). Morpho-Genetic Screening of the Promising Rice Genotypes under Salinity Stress. Journal of Agricultural Science 7(5):94.

Farooq S, Azam F (2002). Molecular markers in plant breeding – 1: concepts and characterization. Pakistan Journal of Biological Sciences 5(10):1135-1140.

Garland SH, Lewin L, Abedinia M, Henry R, Blakeney A (1999). The use of microsatellite polymorphisms for the identification of Australian breeding lines of rice (*Oryza sativa* L.), Euphytica 108:53-63.

Gregorio GB (1997). Tagging Salinity Tolerant Genes in Rice Using Amplified Fragment Length Polymorphism (AFLP). Ph.D. Dissertation. University of the Philippines Los Baños College, Laguna, Philippines P 118.

Hanson B, Grattan SR, Fulton A (1999). Agricultural salinity and drainage. UC DANR Pub 3375. P 160.

Hashimoto Z, Mori N, Kawamura M, Ishii T, Yoshida S, Ikegami M, Takumi S, Nakamura C. (2004). Genetic diversity and phylogeny of Japanese sake-brewing rice as revealed by AFLP and nuclear and chloroplast SSR markers. Theoretical and Applied Genetics 109(8):1586-1596.

Huyen LTN, Cuc L, Ismail Abdelbagi M, Ham LH (2012). Introgression the Salinity Tolerance QTLs *Saltol* into AS996, the Elite Rice Variety of Vietnam. American Journal of Plant Sciences 3:981-987.

Islam MM, Mondol MNH, Emon RM, Begum SN, Bhowmik SK, Hasan AK (2008). Screening of salt tolerant rice genotypes using SSR markers at seedling stage. Bangladesh Journal of Progressive Science and Technology 5(1):45-48.

Islam FSM, Ali Raihan M, Gregorio GB, Islam RM(2012). Genetic diversity analysis of stress tolerant rice (*Oryza sativa* L.). African Journal of Biotechnology 11(85):15123-15129.

Joshi SP, Prabhakar K, Ranjekar PK, Gupta VS (2011). Molecular markers in plant genome analysis. http/www.ias.ac.in/currsci/jul25/articles 15. htm. pp. 1-19.

Kabir MH, Islam MM, Begum SN, Manidas AC (2008). Application of SSR Technique for the Identification of Markers Linked to Salinity Tolerance in Rice. Progressive Agriculture 19(2):57-65.

Lang NT, Buu BC, Ismail A (2008). Molecular Mapping and Marker-Assisted Selection for Salt Tolerance in Rice (*Oryza Sativa* L.). OmonRice 16:50-56.

Lapitan VC, Brar DS, Abe T, Redona ED (2007). Assessment of genetic variability of Philipine rice cultivers carrying good quality traits using SSR Markers. Breeding Science 57:263-270.

Liu K, Muse SV (2005). PowerMarker: Integrated analysis environment for genetic markers data. Bioinformatics 21(9):2128-2129.

Maas EV, Grattan SR (1999). Crop yields as affected by salinity. Agronomy Monograph, Agricultural Drainage. 38:55-108.

Mahalingam A, Saraswathi R, Ramalingam J (2013). Simple sequence repeat (SSR) markers for assessing genetic diversity among the parental lines of hybrid rice (O*ryza sativa* L.). African Journal of

Biotechnology 12(33):5105-5116
Mason AS (2015). SSR Genotyping. In: Batley J, editor. Plant Genotyping. Springer; New York, NY. pp. 77-89.
Ministry of Food and Agriculture (MOFA) (2010). Agriculture in Ghana--Facts and Figures. Statistics, Research and Information Directorate (SRID), Ministry of Food and Agriculture, Accra, Ghana.
Munns R (2002). Comparative physiology of salt and water stress. Plant, Cell and Environment 25:239-250.
Nayar NM (2010). Second Africa Rice Congress, Bamako, Mali, 22–26 March 2010: Innovation and Partnerships to Realize Africa's Rice Potential.
Nwanze KF, Mohapatra S, Kormawa P, Keya S, Bruce-Oliver S (2006). Rice development in sub-Saharan Africa. Journal of the Science of Food and Agriculture 86:675-677.
Perrier X, Flori A, Bonnot F (2003). Data analysis methods. In: Hamon, P., Seguin, M., Perrier X, Glaszmann JC. Ed.,Genetic diversity of cultivated tropical plants. Enfield, Science Publishers. Montpellier pp. 43-76.
Prabakaran A, Paramasivam K, Rajesh T, Rajarajan D (2010). Molecular characterization of rice land races using SSR Electronic. Journal of Plant Breeding 4:512-516.
Ram SG, Thiruvengadam V, Vinod KK (2007). Genetic diversity among cultivars, land races and wild relatives of rice as revealed by microsatellite markers. Journal of Applied Genetics 48(4):337-345.
Roychowdhury R, Joydip K, Malay KA, Narottam D (2013). Physio-Biochemical and Microsatellite Based Profiling of Lowland Rice (*Oryza sativa* L.) Landraces for Osmotic Stress Tolerance. American Journal of Plant Sciences 4:52-63.
Second G (1984). Relations, évolutives chez le genre Oryza et processus de domestication des riz. Etude Set Theses, ORSTOM, Paris, France.
Singh VK, Upadhyay P, Sinha P, Mall AK, Jaiswal SK (2011). Determination of genetic relationships among elite thermosensitive genic Male sterile lines of rice (*Oryza sativa* l.) employing morphological and simple sequence repeat (SSR) markers. Journal of Genetics 90(1):11-19.
Sohrawardy H, Islam MM, Begum SN (2008). Identification of salt tolerant rice lines in F3 population of Pnr-519 x IR -29 using SSR markers. In: Abstracts of Plant Tissue Culture and Biotechnology Conference, 11-13 April, 2008, Dhaka, Bangladesh.
Takeoka T (1965). Taxonomy and chromosome numbers of African representatives of the Oryza officinalis complex. Botanical magazine. (Tokyo) 78:198-201.
Thanh ND, Zheng HG, Dong NV, Trinh LN, Ali ML, Nguyen HT (1999). Genetic variation in root morphology and microsatellite DNA loci in upland rice (*Oryza sativa* L.) from Vietnam. Euphytica 105:43-51.
Thompson MJ, Ocampo M, Egdane J, Rahman MA, Sajise AG, Adorada D L, Tumimbang-Raiz E, Blumwald E, Seraj ZI , Singh RK , Gregorio GB, Ismail AM (2010). Characterizing the Saltol Quantitative Trait Locus for Salinity Tolerance in Rice. Rice 3:148-160.
Wang Y, Buermann W, Stenberg P, Smolander H, Ha¨me T, Tian Y, Hu J, Knyazikhin Y, Myneni RB (2003). Hyperspectral remote sensing of vegetation canopy: Leaf area index and foliage optical properties. Remote Sensing of Environment 85:304-315.
Yang GP, Saghai-Maroof MA, Xu CG (1994). Comparative analysis of microsatellite DNA polymorphism in landraces and cultivars of rice. Molecular and General Genetics 245:187-194.
Zeng L, Taek-Ryoun K, Xuan L, Clyde W, Grieve CM, Gregorio GB (2004). Genetic diversity analyzed by microsatellite markers among rice (*Oryza sativa L.*) genotypes with different adaptations to saline soils. Plant Science 166:1275-1285.
Zeng L, Lesch SM, Grieve CM (2003). Rice growth and yield respond to changes in water depth and salinity stress. Agricultural Water Management 59:67-75.
Zeng L, Shannon MC (2000). Salinity effects on seedling growth and yield components of rice. Crop Science 40:996-1003.

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Evaluation of agronomic performance of beta-carotene rich (yellow fleshed) cassava varieties in Nigeria

Adetoro N. A.[1*], Ogunbayo S. A.[1,2] and Akinwale M. O.[3]

[1]International Institute of Tropical Agriculture (IITA), PMB 5320, Ibadan, Oyo State, Nigeria.
[2]International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), BP 320, Bamako, Mali.
[3]International Institute of Tropical Agriculture (IITA), P. O. Box 30258, Lilongwe, Malawi.

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Twenty-five yellow fleshed cassava varieties including three checks (two improved and one local) were evaluated in a randomized complete block design with four replications in three locations in Nigeria. Tuber yield, dry matter content, root size, fiber content, harvest index, sprouting and vigor of the varieties evaluated were all significant at 0.05 significant levels across the locations. Taste, color of unexpanded leaves, height at branching, and internode length were not significant. In Ibadan, plant height, vigor and root size were not significant. In Mokwa, plant height was not significant. Traits such as dry matter, mealiness and taste were significant. In Onne, dry matter was not significant. Clones such as 01/1413, 01/1442, 01/1663, 98/2132, 01/1277, and 01/1235 were stable across locations, 94/0330 had the highest dry matter (38%) which was better than the best check 30572 (37%). All clones were resistant to cassava mosaic disease, cassava bacteria blight, cassava green mites and cassava anthracnose disease vector infection and to the spread of the pathogens within the plant and across locations. Clones 01/1115, 011413, 01/1663, and 01/1335 had high beta-carotene content of range 7 on a color chart. Clones 01/1368, 01/1371, 98/2132, 90/01554 and 94/0330 had dry matter values ranging from 30 to 38%; these were acceptable values. In terms of yield, the best clones were 01/1368 (26 t/ha), 98/2132 (25 t/ha) and 01/1663 (24.5 t/ha). For gari yield clone 01/1649 gave 25%; 94/0330 gave 23% and 90/01554 gave 23%. They were better than the best check, with 22% garri yield. Cultivating cassava with yellow pigmented root flesh is a valid strategy to solve the problem of improving the nutritional value of the diet in the region where cassava is a staple food.

Key words: Agronomic performance, beta-carotene, clone, evaluation, cassava, yellow cassava, yield.

INTRODUCTION

Cassava (*Manihot esculenta* Crantz) is a major staple food in Nigeria, consumed daily by more than 100 million people. The global production of cassava in 2014 was 278.7 million tons with an estimate of 281 million tons for 2015 and 288.4 million tons for 2016 (FAO, 2016). From available records, Nigeria still stands out as the world's largest producer of cassava with a progressive production pattern that increased from 42.5 million metric tons

*Corresponding author. E-mail: naadetoro@gmail.com.

in 2010 to 54 million metric tons in 2012 with average production output of 12.2 t/ha in 2010 increased to 14.03 t/ha in 2012 (FAOSTAT, 2013). Total area harvested of the crop in 2012 was 3.85 million ha (FAOSTAT, 2013). The diverse uses of cassava largely explain its popularity in the tropics. In Africa, most cassava produced is used for food consumption with 50% in processed form and 38% in the fresh and/or boil form; and 12% is used for animal feed. The diverse uses of cassava largely explain its popularity in the tropics. However, many pathogens and pests reduce cassava yields, especially in Africa including Nigeria. Diseases such as cassava mosaic disease (CMD), transmitted by a whitefly (*Bemisia tabaci*) vector and spread by infected cuttings, cassava brown streak virus disease (CBSD), cassava bacterial blight (CBB; *Xanthomonas axonopodis pv. manihotis*), and anthracnose (CA; *Colletotrichum gloeosporoides*) are among the most important diseases. Pests with a wide African spread are the cassava mealybug (CM; *Phenacoccus manihoti*), African root and tuber scale (*Stictococcus vayssierei*), cassava green mite (CGM; *Mononychellus tanajoa*) and nematodes (particularly *Meloidogyne* species) (Abaca et al., 2014).

Over-dependence on cassava-based diets may result in poor health, stunted growth, reduced capacity for physical activity, and in extreme cases, a high incidence of anaemia, corneal blindness, and compromised immunity (Saltzman et al., 2013). However, while the commonly available white cassava can provide most of the body's daily energy requirements, it does not provide sufficient proteins, essential micronutrients and vitamin A, required for a healthy and productive life. Vitamin A deficiency can impair the body's immunity to infectious diseases and cause eye defect that can lead to partial or complete blindness. Nearly one in three Nigerian children under five and one-quarter of all pregnant women in the country are vitamin A deficient (FAO, 2014). Billions of people around the world suffer from hunger and 'hidden hunger' or micronutrient malnutrition. Around 805 million people were considered chronically undernourished over the 2012 to 2014 period (FAO, 2014).

Those that do not get enough vitamin A and micronutrients (zinc and iron) from the foods they eat face severe health complications and even death. Micronutrient malnutrition can lower intelligence quotient (IQ), cause stunting and blindness in children, lower resistance to disease in both children and adults, and increase risks for both mothers and infants during childbirth. Malnutrition is the underlying cause of 45% of child deaths under the age of 5 (WHO, 2015). In 2013, an estimated 161 children under the age of 5 were stunted (below median height for age) and another 51 million were wasted (below median weight for height) (Thompson et al., 2013).

This is especially true in regions with prolonged dry seasons that limit production and access to alternative sources of micronutrients such as fresh vegetables (Von Grebmer et al., 2014).

Pro-vitamin A varieties that are presently available provide up to 40% of the daily recommended vitamin A intake for children less than 5 years old (De Moura et al., 2015). Nevertheless, new crosses to select varieties with an even higher content of β-carotene varieties are being generated through recurrent selection breeding scheme (Sánchez et al., 2014). This paper reports on the agronomic performance and suitability for quality garri production of adapted beta-carotene rich (Pro-vitamin A) cassava clones in diverse locations (Ibadan, Mokwa, and Onne) in Nigeria.

MATERIALS AND METHODS

The fields were plowed, harrowed and ridged at 1 m apart. Mature stem cuttings (0.25 m long) of 25 genotypes including three checks were planted on plots of four ridges (Table 6). The ridges were about 50 cm high, each 10 m long and spaced 1 m apart. The plot size was 4 m × 10 m (40 m^2). The experimental design was a randomized complete block with four replications. Blocking was done according to the topo-sequence of the field. The plots were weeded six times after planting and no fertilizers were applied. The experiment was conducted in three locations (Mokwa, Ibadan, and Onne) in Nigeria for two seasons (Figure 1). Mokwa (Niger State) is located in the southern Guinea savanna zone with latitude 9°18'N and longitude 5°04'E at about 457 m altitude about sea level (masl) and has a unimodal rainfall pattern with an annual total of 1069 mm, falling between June and October. Radiation is about 450 MJ m^{-2} $year^{-1}$. The soil is alfisols and ultisols. The second environment was Ibadan (Oyo State) with latitude 7°31'N and longitude 3°54'E and is located in the forest savanna transition zone at about 150 masl. It is characterized by a bimodal rainfall also averaging 1300 mm annually, most of which falls between May and October. Radiation is about 5285 MJ m^{-2} $year^{-1}$. The soil is slightly acidic alfisols. The third test environment, Onne (Rivers State), latitude 4° 43'N, longitude 7° 01'E, and 10 masl is in the rainforest zone, has a unimodal rainfall pattern with an annual average of 2400 mm falling between February and December. Relative humidity remains high throughout the year, with average values ranging from 78% in February to 89% in July and September. This site receives an average 4 h of direct sunshine daily, reaching 5060 MJ m^{-2} $year^{-1}$. The soil is representative of highly leached acid ultisols.

Data collection

One month after planting, data was collected on Cassava Mosaic Diseases (CMD). Cassava Bacterial Blight (CBB) was scored monthly until 6 months after planting. Cassava anthracnose disease (CAD) was scored for 6 months after planting and monthly till 9 months after planting. Cassava green mite (CGM) was scored for between January and February. That was when it normally appeared and reached its peak period. The scale used for scoring was 1 to 5 (1= zero attack or resistance; 2 = little attack or little resistance; 3 = medium or moderate resistance; 4 = high attack or susceptible; and 5 = very high attack or highly susceptible).

Number of cassava plants sprouted at 1 MAP was counted and scored as number sprouted or germinated over total number planted.

The plant growth vigor at one month after planting was rated visually, per plot basis, using 3 for low vigor, 5 for intermediate vigor, and 7 for highly vigor cassava plants. Root size was categorized into small, moderate and large with the scale 3 =

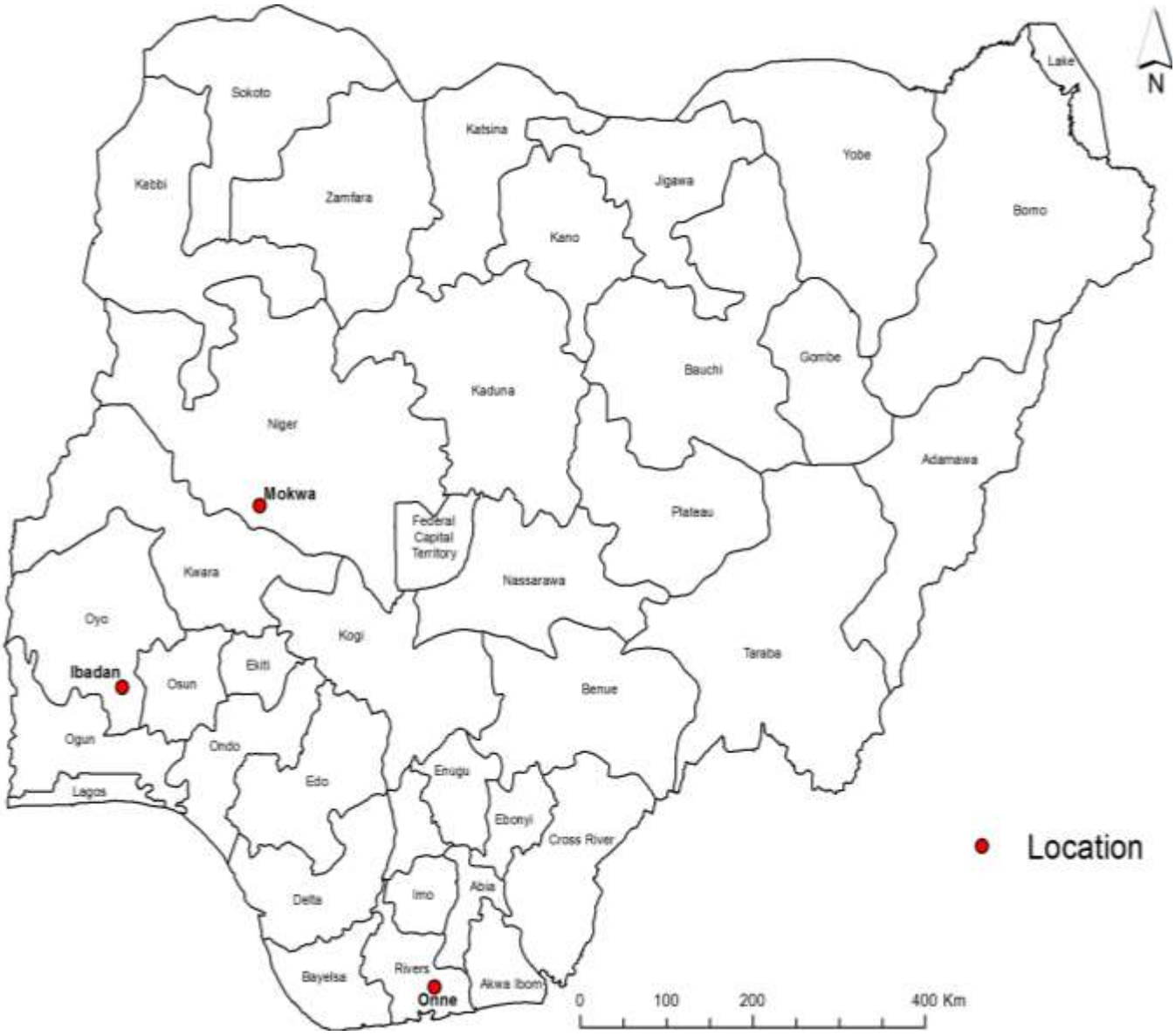

Figure 1. Map of Nigeria showing the three experimental locations.

small, 5 = moderate, and 7 = large. For B-carotene, Provitamin A carotenoids represent precursors to vitamin A in humans. It was scored at harvest with the use of color chart: 1= white, 2= light cream, 3= cream, 4= light yellow, 5= yellow, 6=yellow deep, 7= orange and 8= pink. The root cyanide content was estimated by picrate acid method. It was rated on a 1 to 9 scale based on intensity of red color (higher intensity of higher HCN content of root sample): 1= <10 HCN; 2= 10-15 HCN; 3 = 15-25 HCN; 4 = 25-40 HCN; 5 = 40-60 HCN; 6 = 60-85 HCN; 7 = 85-115 HCN; 8 = 115-150 HCN; and 9 = >150 (Intense red). Taste of boiled roots was examined by panel of five people and the conclusion was recorded. The scale used was 1: sweet, 2: bland, and 3: bitter.

Dry matter content of the tuberous root is an important character for the acceptance of cassava roots by consumers who boil or process them. Fresh sample of 100 g were taken from each clone in each replicate and dried at 70°C in oven and they were re-weighed after 72 h of drying and have attained constant drying. The dried sample was weighed and root dry matter content percentage was calculated as the ratio between fresh weight (FW) and dry weight (DW), that is, DM (%) = (DW/FW) × 100.

For fresh root yield, all the underground roots per plot were weighed in kilogram (kg) and converted to tones per hectare (t/ha).

Garri yield is the weight of garri (a dried granule gotten from 10 kg of fresh cassava roots of each clone, after being peeled, grated, fermented, dewatered, fried and sieved) measured in kilogram.
Harvest index was calculated by (root weight / root weight + shoot weight) × 100.

Statistical analysis

The data collected were subjected to analysis of variance (ANOVA) using General Linear Model (GLM) procedure for randomized complete block design in statistical analysis system (SAS, 1996) to

test for the treatment of effect and significant interaction of the variables considered. The results of the different experiments were subjected to combined analysis of variance to examine genotype by environment interaction (G × E) effect and standard errors were calculated for each trait.

RESULTS AND DISCUSSION

Plant height

Plant height was measured and the mean was calculated. The analysis of variance (Table 4) of the plant height indicated significant differences among the clones at probability level of 0.05 in Onne with CV of 17%, while in Ibadan and Mokwa a non-significant difference was observed.

Vigor

The result of vigor in G × E (Table 5) was very significant with CV of 17.90%. In Ibadan, the CV was 10.66% and not significant. In Onne, the CV was 21% and showed significance and in Mokwa it was 18% and significant. This result shows that vigor is a stable trait and not affected by the environment (Table 4).

Fresh tuber yield

This was very significant among the clones. From G × E analysis, the CV (31.28%) from combined analysis (Table 3) indicated a wide spread of the difference in the mean of the yield among the genotypes and across the locations. The yield could be considered stable at Onne with a high yield of about 24 t/ha. While Ibadan and Mokwa had yield of 15 t/ha (Table 4).

Dry matter

Dry matter content is a very important trait for acceptability of the cassava by consumers. It was significant in Ibadan at 0.05 (Table 4), with CV of 11.17%. In Onne, it was not significant. However, in combined analysis it was significant at a probability level of 0.001 with a CV of 12.88% and has a mean ranged from 28 to 35 across the locations (Table 5). This shows high dry matter percentage. Edoh et al. (2018) also reported higher dry matter percentage of 33.5% in one of her findings.

Cultivating cassava with yellow-pigmented root flesh is a valid strategy to solve the problem of how to improve the nutritional value of the diet in the regions where cassava is a staple food.

From the analysis of the 25 accessions, a significant (p=0.01) carotene content was determined within the range of variability for carotene concentration in the roots. A few genotypes were high up to 7 when a color chart that ranges from 1 to 8 was used. Olapeju et al. (2013) also reported higher carotene concentration in some of the varieties.

Locations accounted for most of the G × E interaction significance (p=0.001) which reflects the differences in soil types in which the clones were grown. This suggested that for the evaluation of cassava clones, it might be more appropriate to test genotypes over space rather than over time. This experiment was able to identify stable clones across locations such as 01/1413, 01/1442, 01/1663, 98/2132, 01/1277, and 01/1235.

Tuber yield, dry matter content, root size, fiber content, harvest index, sprouting and vigor of the varieties evaluated were all significant among the clones in the combined analysis (Table 5). Taste color of unexpanded leaves, height at branching, leaf area, and internode length were not significant. Overall, dry matter content showed that clone 94/0330 (yellow root) had the highest dry matter (38%), which was better than the best check 30572 (37%), followed by the clones 90/01554, 01/1335, 01/1368 and 01/1371 with values ranged from 30 to 35% (Table 3). However, in Ibadan (Table 4), plant height, vigor, mealiness and root size were not significant. In Mokwa (Table 4), sprouting and plant height were the only traits that were not significant. In Onne (Table 4), dry matter, sprouting, mealiness and taste were not significant.

In terms of disease resistance, all the clones evaluated were resistant to CMD and CBB vector infection and to the spread of the pathogen within the plant and across the locations (Tables 1 and 2).

Conclusion

The results showed that all clones were resistant to CMD, CGM and CAD vector infection and to the spread of the pathogen within the plant and across the locations. In terms of beta-carotene, clones 01/1115, 01/1413, 01/1663, and 01/1335 had high beta-carotene content, indicating the best genotypes for recommendation. In terms of dry matter content, clone 94/0330 (yellow root, 38%) was better than the best check 30572 (37%), clones 90/01554, 01/1335, 01/1368 and 01/1371 had dry matter values ranged from 30 to 35%, which were acceptable values. In terms of garri yield; yield per 50 kg of fresh root submitted for garri showed that clones 01/1649 (25%), 94/0330 (23%), and 90/01554 (23%) were better than the check 30572 (22%). Most of the clones evaluated were sweet, some were bland and non-was bitter in terms of taste.

In terms of yield, the best clones were 01/1368 (26 t/ha), 98/2132 and 01/1663 (25 t/ha). Clones 01/1412, 01/1115, 01/1235, 01/1610, 01/1649, and 95/0379 gave between 21 and 22 t/ha. In terms of the cyanide level, clones 01/1442, 01/1413, 01/1115, and 01/1663 were

Table 1. Mean from analysis of variance showing the reaction of 25 yellow root of Cassava genotype to CMD, CBB, CAD and CGM severity and incident at Ibadan, Mokwa and Onne.

Trait	Range	Ibadan			Mokwa			Onne		
		Mean	CV (%)	Sig.	Mean	CV (%)	Sig.	Mean	CV (%)	Sig.
CMD (S)	1-5	1.7	28.8	**	1.42	14.51	**	1.7	15.1	**
CBB (S)	1-5	1.8	22.03	*	2.45	14.31	***	1.49	7.14	*
CMD (I)	1-5	2.02	30.36	Ns	1.11	58.55	***	1.19	59.2	***
CBB (I)	1-5	1.18	39.74	Ns	1.9	18.49	**	1.07	93.58	**
CGM	1-5	4.04	24.6	**	2.9	15.05	**	2.34	6.57	Ns
CAD	1-5	18.8	26.64	Ns	1.89	25.6	ns	1.84	18.29	**

*,**,*** indicate 0.05, 0.01 and 0.001 levels of significance and ns means not significant. CMD: Cassava Mosaic Disease, CBB: cassava bacterial blight, CAD: cassava anthracnose disease, CGM: cassava green mite, S: severity, I: incident. 1: Zero attack or resistance, 2: little attack or little resistance, 3: medium or moderate resistance, 4: high attack or susceptible, and 5: very high attack or highly susceptible.

Table 2. Mean performance of (combined analysis) G × E of beta-carotene in cassava genotypes evaluated for multilocation trial at Ibadan, Mokwa and Onne for disease pest effects.

Trait	Range	Mean	CV (%)	MS between clone (df=24)	Sig. level
CMD (S)	1-5	1.14	21.32	3.2	***
CBB (S)	1-5	1.35	10.64	0.1	***
CMD (I)	1-5	0.28	30.36	0.6	*
CBB (I)	1-5	1.17	39.74	1.6	***
CGM	1-5	3.86	12.2	2.1	***
CAD (S)	1-5	2.11	35.97	0.2	*
CAD (1)	1-5	3.27	36.69	0	***

*,**,*** indicate 0.05, 0.01 and 0.001 levels of significance and ns means not significant. CMD: Cassava Mosaic Disease, CBB: cassava bacterial blight, CAD: cassava anthracnose disease, CGM: cassava green mite, S: severity, I: incident. 1: Zero attack or resistance, 2: little attack or little resistance, 3: medium or moderate resistance, 4: high attack or susceptible, and 5: very high attack or highly susceptible.

Table 3. Mean performance of G × E beta-carotene in cassava genotypes evaluated for multilocational trial at Ibadan, Mokwa and Onne for agronomic traits effect.

Clone	Fresh yield	H.I.	DM (%)	Beta carotene	Cyanide	Root size	Taste	Garri yield/50 kg	Garri yield
01/1115	21.21	1.2	29.53	7	3.5	7	1.8	8	16
01/1224	17.96	0.5	34.17	6.8	4.75	7	1.5	10.5	21
01/1235	21.4	0.59	28.99	6	4.5	7	1.3	6	12
01/1273	16.21	0.5	28	6.8	4.75	7	1.7	7.2	14.4
01/1277	16.2	0.5	34.12	6.5	4.5	6.3	1.7	7.5	15
01/1331	9.28	0.36	30.84	6.3	5.5	5.3	2	7.3	14.6
01/1335	18.5	0.53	31.88	7	4.25	7	1.5	8.5	17
01/1368	26.13	0.05	30.12	6	5.25	7 .0	1.8	7	14
01/1371	17.86	0.52	30.04	6.8	4.5	6.7	2	6.5	13
01/1412	21.96	0.58	28.08	6.5	4.5	7	1.8	10	20
01/1413	19.06	0.52	28.97	7	3.25	5.8	1.5	2	4
01/1442	16.58	0.53	30.44	6.3	3	6.3	1.7	7	14
01/1610	20.61	0.52	27.52	6.8	4.75	6.7	1.8	10	20
01/1646	18.1	0.45	31.58	5.5	4	6.3	1.7	7.7	15.4
01/1649	20.88	0.56	32.19	6.3	4.25	7	1.5	12.5	25
01/1662	16.38	0.46	29.9	5.5	4	6.5	1.8	10.6	21.2
01/1663	24.54	0.54	29.02	7	3.5	7	2	10	20
30572	26.83	0.55	37.18	1	3.75	6.7	1.5	11	22
90/01554	19.95	0.49	34.97	4.3	4.25	7	1.8	11.5	23

Table 3. Contd.

91/02324	24.66	0.6	35.3	1	3.25	7	1.3	8.2	16.4
94/0006	20.84	0.6	35.19	4.3	4.25	6.7	1.7	7.4	14.8
94/0330	13.89	0.41	38.35	4.5	5.25	6.3	1.7	11.5	23
95/0379	20.81	0.55	29.59	6	4	6.7	1.8	6.5	13
98/2132	25.02	0.69	35.78	6	4.25	7	2	7	14
TME 1	18.01	0.54	33.62	1	3.25	6.8	1.5	8.2	16.4
G.MEAN	19.73	0.55	31.81	5.5	4.23	6.68	1.7	8.4	-
STDEV	3.91	0.15	3.09	1.9	0.67	0.43	0.2	2.23	-
Stderr	0.01	0.03	0.62	0.4	0.13	0.12	0.06	0.64	-
CV%	31.28	0.6	12.88	7.5	2.69	12.49	12.9	9.78	-
F-Ratio	***	***	***	***	***	***	ns	ns	-

*,**,*** indicate 0.05, 0.01 and 0.001 levels of significance and ns means not significance. H.I: Harvest index, DM: Dry matter

Table 4. Mean square from analysis of variance showing various agronomic traits of 25 yellow root cassava genotypes evaluated for multilocational trail at Ibadan, Mokwa and Onne.

Trait	Ibadan				Mokwa				Onne			
	Mean	CV (%)	MSBC (Df=24)	SL	Mean	CV (%)	MSBC (df=24)	SL	Mean	CV (%)	MSBC (Df=24)	SL
FYLD	15.2	35.8	86.83	**	15.1	42	258.4	***	24.1	25	116	***
Sprout	95.67	7.79	337.1	*	18.16	66	221.6	Ns	93.9	9.4	205	Ns
Vigor	6.25	10.66	1.78	Ns	5.64	18	1.88	**	4.4	21	1.7	*
LA	11697	228.5	6736	Ns	15222	58.4	54428	Ns	26598	66.4	46798	Ns
Plant H.	63.26	41.28	428	ns	59.7	30	277	Ns	117	17	695	*
DM	35.26	11.17	37.4	Ns	34.5	15	34.56	*	28	18	35	Ns
Mealy	1.05	119.7	2.5	Ns	2.64	14	0.53	***	0.5	131	0.82	Ns
Cyanide	5.68	20.47	3.22	**	5.68	20	3.22	***	4.2	20	4.21	*
B.carotene	5.45	9.88	10.55	**	5.43	9	11.61	**	4.6	19	3.23	*
Rt. size	6.68	12.06	0.57	Ns	6.64	11	1.38	**	6.6	13	2.6	***
Taste	1.65	24.63	0.44	**	1.68	20	1.06	*	1.8	27	0.3	Ns
Garri	20.65	15.23	20.68	**	19.65	24	15.65	*	19	9.8	19	***

*, **,*** indicate 0.05, 0.01 and0.001 levels of significance and ns means not significance. FYLD: Fresh yield, LA: leaf area, Plant H.: plant height, DM: dry matter, B.carotene: beta carotene, Rt. size: root size, MSBC: mean square between clones, SL: significant level.

Table 5. Mean performance of G × E of 25 beta-carotene cassava evaluated for multilocational trail at Ibadan, Mokwa and Onne for agronomic traits effect.

Trait	Mean	CV (%)	MS between clone (df=24)	Sig. level
Fresh yield	19.63	31.28	125	***
Sprout	1.92	11.19	0.06	***
Vigor	9	17.9	3.03	***
H.I	0.51	14.55	0.02	***
Root size	6.63	12.49	1.74	***
Fiber	2.37	9.36	0.16	***
Dry matter	32.91	12.88	50.1	***
Taste	69	25.09	0.24	Ns
Leaf shape	13	12.03	1.17	***
Color of unexpanded leaf	65	34.65	11.47	Ns
Pubescent of young leaf	2.78	45.77	5.5	**

Table 5. Contd.

Petiole length	12.33	34.36	35.21	*
Petiole Color	3.5	29.37	6.5	***
Flowering	1.7	35.79	0.31	**
Fruit	1.2	89.35	0.37	*
Height at branching	26.91	81.5	799.9	ns
Stem/plant	1.19	27.31	0.44	**
Internode length	1.21	18.87	0.05	Ns
Stem color	2.5	23.85	1.26	**
D. of anthocyanine pigment	1.81	60.62	6.64	*

*,**,*** indicate 0.05, 0.01 and 0.001 level of significance and ns mean not significant.

Table 6. Characteristics of 25 yellow root cassava clones evaluated for agronomic performance during 2003/2004 cropping season.

Clone	Leaf length (cm)	Color of unexpanded leaf	Petiole length (cm)	Plant height (cm)	Petiole color	Flower	No. of stem Plant^{-1}	Internode length (cm)	Stem	Height at branch (cm)
01/1115	10.7	Light-green	12.7	91.5	Light-green	Present	2	1.5	Silver-green	25
01/1224	13.8	Green-purple	18.2	105.5	Dark-green	Present	2	2	Dark-green	26.9
01/1235	11.2	Green-purple	27.1	68.3	Dark-green	Present	3	2	Silver-green	59.6
01/1273	11.1	Green-purple	16.5	69.5	Light-green	Present	2	1.5	Light-brown	40
01/1277	14.6	Purple	20.9	122.8	Light-green	Absent	3	2	Light-brown	73.2
01/1331	15.2	Green-purple	14.7	105.1	Light-green	Present	2	2	Dark-brown	35.2
01/1335	13.5	Green-purple	13.4	120.3	Dark-green	Present	2	2	Dark-green	26.8
01/1368	12.8	Green-purple	18.9	127.2	Light-green	Present	3	2	Light-brown	23.2
01/1371	8.4	Green-purple	15	131.3	Light-green	Present	2	2	Dark-green	54
01/1412	12.2	Dark-green	17.8	112.9	Light-green	Present	3	2	Silver-green	22
01/1413	12.5	Green-purple	17	108.2	Dark-green	Present	3	1.5	Silver-green	16.3
01/1442	13	Dark-green	12	130.1	Light-green	Present	3	2	Dark-green	23.3
01/1610	14	Dark-green	18.8	97	Light-green	Present	3	1.5	Light-brown	25
01/1646	14.7	Green-purple	24.1	143.9	Light-green	Absent	2	2	Light-brown	29.8
01/1649	14.2	Purple	18.3	117.9	Light-green	Present	2	2	Light-brown	55.8
01/1662	15.8	Dark-green	20.5	142	Dark-green	Present	2	2	Dark-green	31.3
01/1663	15	Dark-green	25.2	141.4	Dark-green	Absent	3	1.5	Dark-brown	0
30572	10.4	Green-purple	24.2	62.3	Dark-green	Present	2	2	Dark-brown	74.8
90/01554	15.7	Green-purple	17	104.9	Dark-green	Present	3	2	Dark-green	21.9
91/02324	15.2	Green-purple	25.3	87.6	Dark-green	Absent	2	2	Dark-brown	0
94/0006	13	Light-green	19.8	112.4	Dark-green	Present	2	2	Dark-brown	30
94/0330	13	Purple	23.7	125.5	Red	Present	2	1.5	Dark-brown	31
95/0379	14.3	Dark-green	19	128.3	Dark-green	Present	1	2	Dark-brown	45
98/2132	10.2	Green-purple	23	60	Dark-green	Present	2	2	Dark-brown	25
TME 1	16	Purple	26	250	Purple	Absent	1	1.5	Dark-brown	150

very low, clones 01/1224, 01/1235, 01/1371, 95/0379, 98/2132, 94/0006, 01/1662, and 01/1412 were moderate.

None of them was high in cyanide level. Most of the root sizes were large and some were moderate while none were small among the clones evaluated.

In terms of harvest index, clone 01/1115 had the highest index of about 120% of the total yield. Clones 98/2132 (69%), 01/1235 (59%), 01/1412 (58%), and 95/0379 (55%) were acceptable. Clones 01/1115 and 98/2132 were better than the best check (91/02324) with a harvest index of 60%. Clones 01/1235, 01/1412, 98/2132, 01/1115, 94/0006, and 01/1649 were better than the most popular check 30572 (55%).

Four of the clones used in this experiment were already released varieties in Nigeria. Three of them (01/1368, 01/1412 and 01/1371) were released in the year 2011,

while one (98/2132) was released in the year 2012.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abaca A, Kiryowa M, Awori E, Andema A, Dradiku F, Moja AS, Mukalazi J (2014). Cassava Pests and Diseases Prevalence and Performance as Revealed by Adaptive Trial Sites in North Western Agro-Ecological Zone of Uganda. Journal of Agricultural Science 6(1):116-122.

De Moura FF, Moursi M, Lubowa A (2015). Cassava intake and Vitamin A status among women and preschool children in Akwa-Ibom, Nigeria. PLoS One 10(6):e0129436.

Edoh NL, Adiele J, Ndukwe I, Ogbokiri H, Njoku DN, Egesi CN (2018). Evaluation of High Beta Carotene Cassava Genotypes at Advanced Trial in Nigeria. Biological Sciences, Chemical Sciences, Physical Sciences, Medicine, Engineering & Technology. The Open Conference Proceedings Journal ISSN: 2210-2892 -Volume 9, 2018.

Food and Agriculture Organization of United Nations (FAOSTAT) (2013). Production, Crops, Cassava 2012 dataFood http://www.fao.org/faostat/en/#data/QC

Food and Agriculture Organization of the United Nations (FAOSTAT, 2014). The State of Food and Agriculture: Innovation in Family Farming FAO, Rome.

Food and Agriculture Organization of United Nations (2016). Food outlook, Biannual report on global markets, market summaries: World production of cassava. FAO.5.

Olapeju O, Phorbee, Ibiyemi O, Olayiwola S, Sanni A (2013). Bioavailability of Beta Carotene in Traditional Fermented, Roasted Granules, *Garri* from Bio-Fortified Cassava Roots Food and Nutrition Sciences, 2013, 4:1247-1254 Published Online December 2013. http://www.scirp.org/journal/fns

Saltzman A, Birol E, Bouis HE, (2013). Biofortification: progress toward a more nourishing future. Global Food Security 2(1):9-17.

Sánchez T, Ceballos H, Dufour D (2014). Prediction of carotenoids, cyanide and dry matter contents in fresh cassava root using NIRS and Hunter colour techniques. Food Chemistry 151:444-451.

Thompson A, Blossner M, Borghi E, Feng J, Mistiaen J, (2013). Levels and Trends in Child Malnutrition, UNICEF-WHO-The World Bank, Joint Child Malnutrition Estimates, Available from: <http://www.who.int/nutgrowthdb/summary_jme_.

Von Grebmer K, Saltzman A, Birol E, Wiesmann D, Prasai N, Yin S, Yohannes Y, Menon P, Thompson J, Sonntag A (2014). Global Hunger Index: The Challenge of Hidden Hunger. Welthungerhilfe, International Food Policy Research Institute, and Concern Worldwide, Bonn, Washington, D.C., and Dublin, 50. http://dx.doi.org/10.2499/9780896299580.

World Health Organization (WHO) (2015). Global Database on Child Growth and Malnutrition, Available from: http://www.who.int/nutgrowthdb/about/en/

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Genetic variability, heritability and genetic advance of maize (*Zea mays* L.) inbred lines for yield and yield related traits in southwestern Ethiopia

Tadesse Jilo[1], Leta Tulu[2], Techale Birhan[3] and Lemi Beksisa[4*]

[1]Bonga University, College of Agriculture and Natural Resource, Department of Plant Science, Bonga, Ethiopia.
[2]National Agricultural Biotechnology Research Center, P. O. Box 31, Holeta, Ethiopia.
[3]Jimma University, College of Agriculture and Veterinary Medicine, P. O. Box 370, Jimma, Ethiopia.
[4]Jimma Agricultural Research Center, P. O. Box 192, Jimma, Ethiopia.

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Understanding the genetic variability, heritability and genetic advance of traits in any plant population is an important pre-requisite for breeding program. The experiment was conducted to assess the magnitude of genetic variability, heritability and genetic advance of 24 maize inbred lines for 16 quantitative traits. The field experiment was conducted during 2016 cropping season at Jimma Agricultural Research Center (JARC). Alpha lattice (0, 1) design with three replications and nine blocks was used. Analysis of variance showed high significance (P<0.01) differences among genotypes for all traits studied except tassel size. The genotypic coefficient of variation (GCV) for all traits studied was smaller than the phenotypic coefficient of variation (PCV), indicating the significant role of environment in the expression of traits studied. The estimates of PCV and GCV was high for grain yield, thousand kernel weight, ear height, ear diameter, anthesis and silking interval and plant aspect. Heritability estimates ranged from 9.15 for tassel size to 96.02 for thousand kernel weight. Estimates of genetic advance as percent of mean at 5% selection intensity ranged from 2.76% for days to maturity to 50.69% for grain yield. High heritability along with high genetic advance was obtained for plant height, ear length and 1000-kernel weight, indicating the predominance of additive gene effects in controlling the traits and effective selection on the basis of these traits would be absolutely useful for the improvement of inbred lines. Therefore, it could be recommended that due emphasis should be given for these traits for the improvement of maize inbred lines.

Key words: Heritability, genetic variability, genetic advance, inbred lines.

INTRODUCTION

Maize (*Zea mays* L, 2n=2x=20), a member of the grass family Gramineae (Poaceae), is one of the oldest cultivated crops. Maize is predominately cross pollinated by wind, but self-pollination is also possible (Sleper and Poehlman, 2006). Maize is the most important crop worldwide and basic trade product recurring ingredient for millions of people in Sub-Saharan Africa (Nzuve et al., 2013). Maize has also become the most

*Corresponding author. E-mail: lbeksisa@gmail.com. Tel: +251910822464.

most important staple food in rural Ethiopia (Abate et al., 2015). In addition, maize is also gaining popularity in Ethiopia for its demand for stover as animal fodder and a source of fuel for rural families. In Ethiopia, maize as food constitutes about 20% consumption and constitutes about 13% as feed in total cereals (FAO, 2013). It contributes about 29% of the calorie intake from total cereal consumption, followed by wheat and teff which is contributing about 21 and 17%, respectively (FAO, 2013).

Maize is the global leading cereal in terms of annual production which is about 1040.21 million tons, followed by wheat with production of 748.24 million tons and third in area under cultivation among all cereal crops. In Ethiopia, maize ranks second after teff in area coverage and first in total production (Abate et al., 2015). In Ethiopia, maize grows under a wide range of environmental conditions from 500 to 2400 m.a.s.l. The mid- altitude, sub-humid agro-ecology is, however, the most important maize producing environment in the country (Kebede et al., 1993). Though Ethiopia compares favorably with the main maize producing country in Sub Saharan Africa, the country is yet to achieve its potential in terms of production because of the old varieties dominating the seed system in the country, many biotic and abiotic factors, lack of genetically diverse source materials and little success in developing high yielding hybrids and synthetic varieties for different agro ecologies of the country (Abate et al., 2015). In particular, most of the varieties grown in southwestern highland of the Ethiopia are low yielding local cultivars with very tall plant ear, resulting in root and stalk lodging.

Information on the nature and magnitude of variability and heritability in a population is one of the prerequisites for successful breeding program in selecting genotypes with desirable characters (Dudly and Moll, 1969). Genetic improvement in traits of economic importance along with maintaining sufficient amount of variability is always the desired objective in maize breeding programs. In order to improve the genetic diversity of local germplasm, it is important to know the extent of already existing genetic variations in the material. The productivity and quality of maize are assured through initially evaluating, identifying and properly selecting of promising parental lines from available maize inbred lines.

In Ethiopia, maize improvement started half a century ago (Mosisa et al., 2002). During the late 1960s and early 1970s, several promising genotypes of east African origin were introduced and evaluated at different locations. Different studies were conducted to elucidate the nature and magnitude of genetic variability among maize inbred lines which identified traits like ear length, ear diameter, kernel number per row, ears per plant, 100 seed weight and rows per ear as potential selection criteria in a breeding program. For instance, Aman et al., (2015) identified traits such as kernels row per ear, grain yield, kernels number per row, 1000-kernel weight, ear diameter, ear length, days to anthesis, days to silking, plant height, ear height, days to maturity and plant aspect as important traits in maize improvement. The national maize breeding program of Ethiopia also developed several maize inbred lines for use. However, little effort has been made; particularly in mid altitude area of the country to determine the variability of maize inbred lines considering their different morphological traits. Thus, the study was undertaken to determine the nature and magnitude of genetic variability among maize inbred lines.

MATERIALS AND METHODS

Description of the study site

The study was conducted at the Jimma Agricultural Research Center (JARC) during the main cropping season of 2016. The center is located in Ethiopia, Oromia National Regional State in Jimma zone, 343 km to the southwest of the capital city of the country, Addis Ababa. It is located around 07°46'N latitude and 36°47'E longitude coordinate and at an elevation of 1753 m.a.s.l. It represents the mid-altitude agro ecological zones which receive an annual rainfall of 1607.99 mm, with minimum and maximum mean temperatures of 16.60 and 31.48°C, respectively (JARC Agro-metrology department, 2016). The major soil type of the area is Chromic Nitosol and Combisol of upland and fluvisol of bottom land with a pH of about 5.2 (EIAR, JARC profile).

Experimental materials

The experimental materials were obtained from Bako National Maize Research Center (BNMRC). Twenty-four commercial inbred lines were used. Description of the experimental materials used in this study is indicated in Table 1.

Experimental design and field management

The treatments were evaluated in alpha lattice design (Patterson and Williams, 1976) in three replications. Each treatment was planted in four rows of 5.1 m length with spacing of 0.75 m between rows and 0.30 m between plants within the rows. Two seeds were planted per hill and then thinned to one plant per hill to achieve standard plant density of 44,444 plants per hectare. The middle two rows were used for observations and data recording. Field management and other agronomic management practices were done following research recommendations for the area.

Data collected

Data were recorded on individual plant and plot bases for sixteen quantitative traits at the appropriate growth stage of the crop depending on the standard evaluation system of maize plant (IBPGR, 1991).

Data recorded on a plant basis

Plant height (cm)

The height of the ten randomly selected plants was measured from base of the plant to the base of the first branch of the tassel and the average of the ten plants recorded as plant height for the plot.

Table 1. List of inbred lines used in the study.

Line No	Entry name	Source
1	124-b(109)	BNMRC
2	142-1-e	BNMRC
3	144-7-b	BNMRC
4	35B-190-0-S10-2-1-2-2-1-2	BNMRC
5	A-7033	BNMRC
6	BKL001	BNMRC
7	BKL002	BNMRC
8	BKL003	BNMRC
9	CML124-b(113)	BNMRC
10	CML159	BNMRC
11	CML144	BNMRC
12	CML161	BNMRC
13	CML165	BNMRC
14	CML197	BNMRC
15	CML202	BNMRC
16	CML312	BNMRC
17	CML334	BNMRC
18	CML395	BNMRC
19	CUBA	BNMRC
20	F-7215	BNMRC
21	ILOO,EI-1-9-1-1-1-1-1	BNMRC
22	MBRC5BCF108-2-3-1-B-5-2-B-B-B-B	BNMRC
23	PO,OOE3-2-1-2-1	BNMRC
24	SC22	BNMRC

Source: Bako National Maize Research Center.

Ear height (cm)

The height of the ten randomly selected plants was measured from base of the plant to the node bearing the upper most ear of the same plant used to measure plant height and the average was recorded as ear height for the plot.

Leaf length (cm)

Leaf length was measured from five randomly selected leaves per plant in the leaf subtending the upper ear from the ligule to apex for all ten randomly selected plants after anthesis and the average was recorded for the plot.

Leaf width (cm)

Leaf width was measured for five randomly selected leaves per plant in the leaf subtending the upper ear at the widest point along its length for all ten randomly selected plants after anthesis and the average was for the plot.

Leaf area (cm^2)

Is the average area of five randomly selected leaves per plant in the plot as the product of its length and width taken from each of 10 sampled plants, then multiplied by the correction factor k (k=0.75), where LA= LW x LL x K.

Ear length (cm)

The length of ears harvested from the ten randomly selected plants was measured using ruler and the average was recorded for the plot. The same ears used to record ear diameter were used to record ear length.

Ear diameter (cm)

The diameter of ears harvested from the ten randomly selected plants was measured using Digital caliper and the average was recorded for the plot.

Number of kernel rows per ear

The total number of rows was counted in ten randomly taken ears and the average value was recorded as number of rows per ear.

Data recorded on plot basis

Days to 50% anthesis

Days to anthesis was recorded as the number of days from planting

to the day when 50% of the plant in a plot started pollen shading.

Days to 50% silking

Days to silking was recorded as the number of days from planting when 50% of plants in the plot had their silks emerged 2-3 cm above the sheath.

Anthesis and Silking interval

Anthesis-silking interval was determined as the difference between the numbers of days to anthesis and silking.

Tassel size

Tassel size was recorded after milk stage using 3-7 scale; where 3, 5 and 7 indicate small, medium and large, respectively.

Plant aspect

Plant aspect was scored using 1-5 scale, where 1-indicates good (considering ear size, uniformity, disease infestation, husk cover, and so on) while 5 indicates poor genotype having undesirable ear characters.

Days to maturity

Days to maturity was recorded as the number of days from planting to the day on which 50% of the plants in the plot formed a black layer on sampled grains.

Thousand kernel weight (g)

After shelling, random sample of kernels from the bulk of each experimental unit was counted using a photoelectric seed counter and weighed in grams on sensitive balance after the moisture has been adjusted to 12.5%.

Grain yield (t/ha)

Field weight of all harvested ears was weighted and converted to grain yield using shelling percent of 80 percent. Grain yield was then determined in tons per hectare after adjusting moisture content of 12.5 percent using the following formula.

Grain yield =fresh weight $\times[\frac{\mathbf{100-moisture}}{\mathbf{100}}]\mathbf{x}\left[\frac{\mathbf{1.176x0.8x100}}{\mathbf{plot\ size\ in\ m^2}}\right]$ Kidistet al. (2013)

0.8 is shelling ratio, while 1.176 is constant.

Data analysis

The data collected for each character were subjected to analysis of variance (ANOVA) using statistical software, SAS version 9.3.

Estimation of variance components

Variance components were estimated to identify genetic variability among inbred lines. Error (σ^2e), genotypic (σ^2g) and phenotypes (σ^2p) variances were calculated from expected mean squares of analysis of variance by adopting the formula suggested by Hallauer and Miranda (1988).

Error variance

$\sigma^2e = MSe$

Where: MSe= mean square of error

Genotypic variance

$\sigma^2g = (\frac{MSg-MSE}{r})$

Where: MSg= mean square of genotype, MSe= mean square of error and r=number of replications

Phenotypic variance

$\sigma^2p= \sigma^2e+ \sigma^2g$

Where: σ^2e= error variance and σ^2g=genotypic Variance

Phenotypic and genotypic coefficients analysis

Genotypic coefficient of variation (GCV)

$GCV = (\frac{\sqrt{\sigma^2g}}{\bar{x}})100$

Where: σ^2g= genotypic variance and $\bar{x}$= mean of the trait

Phenotypic coefficient of variation (PCV)

$PCV = (\frac{\sqrt{\sigma^2p}}{\bar{x}})100$

Where: σ^2p= phenotype variance and $\bar{x}$= mean of the trait

PCV and GCV values were categorized as low, moderate, and high values as indicated by Sivasubramanian and Menon (1973) as follows:

0-10% = low, 10-20% = Moderate and >20% = high

Heritability and genetic advance estimation

Heritability broad sense (H^2) = $(\frac{\sigma^2g}{\sigma^2p})100$

Where: σ^2g= genotypic variance and σ^2p= phenotypic variance.

Then it was categorized as low, moderate and high as indicated by Robinson et al. (1955).

0-30% =low**,** 30-60% =moderate and >60 =high

Genetic advance (GA) and genetic advance as per cent of the mean (GAM) were estimated as devised by Johnson et al., (1955), that is,

$GA = k\sigma pH^2$

Where: σp= the phenotypic standard deviation of the character**,** H^2= broad sense heritability estimate and k = selection differential where k=2.063 at 5 % selection intensity.

Genetic advance as per cent of the mean was calculated to compare the extent of predicted advances of different traits under selection.

GAM = $(\frac{GA}{\bar{x}})$ 100 (Falconer, 1996).

Table 2. Mean squares of variance for different studied traits in maize inbred lines.

Traits (Source of variation)	Replication df(2)	Block df(6)	Genotype df(23)	Error df(46)	CV	R.E
MFD	16.79	3.72	49.82**	6.37	2.88	1.14
FFD	20.68	3.42	45.24**	5.95	2.70	1.15
ASI	0.26	1.24	1.15*	0.58	28.82	0.58
LL	95.37	5.44	116.29**	11.75	4.30	1.10
LW	11.29	1.64	4.58**	0.705	6.97	1.10
LA	81095.07	7544.69	24528.62**	3879	8.65	1.01
TS	6.05	3.62	2.72^{ns}	2.09	28.58	0.94
PH	3842.54	271.43	1829.73**	233.7	8.73	1.18
EH	3257.10	95.04	924.81**	211.0	15.76	1.11
PA	1.05	0.32	1.05**	0.42	22.80	1.51
MD	13.72	4.62	36.76**	9.17	1.89	1.22
ED	344.95	344.95	250.40**	110	26.95	1.10
EL	37.24	4.87	12.54**	1.94	10.24	1.10
TKW	0.001	0.002	0.012**	0.001	3.97	1.0
KRPE	2.59	1.66	3.13**	1.29	8.53	1.30
KNPR	72.42	44.88	52.93**	14.30	14.84	1.46
GY	592.16	163.11	314.92**	60.39	27.12	1.25

ASI=Anthesis and Silking Interval, CV=Coefficient of Variation, ED= Ear Diameter, EH=Ear Height, EL=Ear Length, FFD=Days to 50% silking, GY=Grain Yield, KRPE=Kernel row per ear, LA= Leaf Area, LL=Leaf Length, LW=Leaf Width, MD=Maturity Date, MFD=Days to 50% anthesis, NKPR=Number of kernel per row, PA=Plant Aspect, PH=Plant height, R.E=Relative efficiency, TKW=Thousand Kernel Weight, TS=Tassel Size.

GA = genetic advance, and $\bar{x}$= mean for the trait
The GA as percent of the mean was categorized as low, moderate and high according to Johnson et al. (1955) as follows.

0-10% =low**,** 10-20% =moderate and >20% =high

RESULTS AND DISCUSSION

Analysis of variances

There were highly significant (P≤0.01) differences among the inbred lines for all studied traits, except for tassel size (Table 2). The present result is also in agreement with the findings of Taye (2014) and Mustafa et al. (2015)**.** Thus, the genetic variability recorded in this study could be exploited by plant breeders to develop improved hybrid varieties.

Mean and range values

The range and mean values for the 16 traits are presented in Table 3. The results indicated significant differences among the inbred lines for growth, yield and yield related traits. The mean grain yield ranged from 5.69 to 52.21 ton per hectare. Among the studied inbred lines 45.83% of inbred lines gave above the grand mean. Number of kernels per row ranged from 16.07 to 34.1307 with a mean value of 25.48 numbers. The range observed for kernel row per ears was 10.93 to 15.33 with overall mean of 13.31 rows. The maximum and minimum values of 1000-kernel weight were 0.43 and 0.20 t/ha respectively, with a mean value of 0.32t/ha.

The mean plant and ear heights of the genotypes ranged from 136.33 to 235.67 cm and 70.00-144.67 cm respectively. The range observed for days to 50% silking was 82.67 to 100.67 days, with overall mean of 90.28 days. Days to 50% anthesis varied from 82.67 to 100.67, with overall mean of 87.62 days. Days to maturity varied from 155 to 167.67 days, with a mean of 160.44 days. Among total studied inbred lines 58.33% of inbred lines were indicated to exhibit early maturing day.

The inbred lines CML359, CML144 and PO, OOE3-2-1-2-1 were found to be superior in terms of grain yield per hectare, as well as in other important yield components. It is, therefore, suggested that these lines could be used for further improvement of maize inbred lines for yield targeted breeding strategy. Particularly, the wide variability observed for grain yield as a quantitatively inherent character among the genotypes means that, there is ample opportunity for selection in the genotypes for improvement of this important economic character. The range and mean values of the studied traits suggested the existence of sufficient variability among the studied inbred lines for the majority of considered characters and their considerable potential for improvement. Tulu et al. (2014) reported a wide range of variability of traits such as grain yield per hectare, row

Table 3. Components of variance, coefficients of variability (%), heritability (H^2), expected genetic advance (GA) and genetic advance as percent of mean (GAM).

Traits	Range	Mean ± SD	(σ^2g)	(σ^2p)	(σ^2e)	GCV%	PCV%	H^2%	GA	GAM%
MFD	82.67-100.67	87.62 ± 3.81	14.48	20.85	6.37	4.34	5.21	69.46	6.54	7.47
FFD	85.33-103.00	90.28 ± 3.62	13.11	19.05	5.95	4.01	4.83	68.75	6.19	6.86
ASI	1.33-4.00	2.65 ± 0.43	0.19	0.77	0.58	16.39	33.16	24.44	0.44	16.72
LL	64.00-95.67	79.71 ± 5.90	34.85	46.59	11.74	7.41	8.56	74.78	10.53	13.21
LW	10.00-14.67	12.04 ± 1.14	1.29	1.99	0.70	9.44	11.74	64.70	1.89	15.66
LA	580.50-956.00	720.1 ± 82.96	6882.0	10766.1	3879.6	11.52	14.40	63.95	136.87	19.01
PH	136.3-235.6	175.2 ± 23.06	531.11	765.76	233.77	13.16	15.79	69.47	39.66	22.63
EH	70.00-144.67	92.19 ± 15.42	237.92	448.96	211.04	16.73	22.98	52.99	23.16	25.13
PA	2.00-4.67	2.85 ± 0.46	0.21	0.63	0.42	16.14	27.94	33.38	0.55	19.24
MD	155-167.67	160.44 ± 3.03	9.19	18.36	9.17	1.91	2.67	50.07	4.42	2.75
ED	20.44-50.7	38.92 ± 6.84	46.77	156.85	110.08	17.57	32.17	29.81	7.70	19.80
EL	10.53-19.27	13.59 ± 1.89	3.53	5.47	1.93	13.83	17.21	64.61	3.12	22.94
TKW	0.20-0.43	0.317 ± 0.06	0.003	0.003	0.001	19.51	19.91	96.02	0.12	39.44
KRPE	11.3-15.3	13.31 ± 0.78	0.61	1.90	1.29	5.88	10.36	32.25	0.91	6.90
KNPR	16.1-34.3	25.48 ± 3.59	12.87	27.17	14.30	14.08	20.46	47.37	5.09	19.99
GY	5.69-52.21	28.65 ± 9.21	84.84	145.24	60.39	32.15	42.06	58.42	14.52	50.69

ASI=Anthesis and Silking Interval, ED= Ear Diameter, EH=Ear Height, EL=Ear Length, FFD= Days to 50% silking , GY=Grain Yield, KRPE=Kernel row per ear, LA= Leaf Area, LL=Leaf Length, LW=Leaf Width, MD=Maturity Date, MFD=Days to 50% anthesis, NKPR=Number of kernel per row, PA=Plant Aspect, PH=Plant height, TKW=Thousand Kernel Weight,

number per ears, number of kernels per row, ear length, ear diameter and 1000-kernel weight which is confirmed by the present study. Iqbal et al. (2015) also observed high range values for plant height and ear height in maize inbred lines they studied.

Genotypic and phenotypic coefficients of variation

Estimated variance components of PCV and GCV of the studied traits are presented in Table 3. High GCV was observed for grain yield (32.15). On the contrary, moderate values were observed for thousand kernel weight (19.51), ear height (16.73), ear diameter (17.57), anthesis and silking interval (16.39), plant aspect (16.14), number of kernels per row (14.08), ear length (13.83), plant height (13.16) and leaf area (11.54). The rest of the traits such as, leaf width (9.44), leaf length (7.41), kernel row per ear (5.88), days to 50% anthesis (4.34), days to 50% silking (4.01) and days to maturity (1.91) depicted low genotypic coefficient of variation.

High PCV was observed for grain yield (42.06), anthesis and silking interval (33.16), ear diameter (32.17***)***, plant aspect (27.94) and ear height (22.98). On the contrary, moderate values were observed for number of kernels per row (20.46), thousand kernel weight (19.91), ear length (17.21), plant height (15.79), leaf area (14.40) and leaf width (11.74). The remaining traits such as, number of kernel rows per ears (10.36), leaf length (8.56), days to 50% anthesis (5.21), days to 50% silking (4.83) and days to maturity (2.67) showed a low phenotypic coefficient of variation.

A high range of PCV and GCV was noted for grain yield per hectare, anthesis and silking interval, ear diameter, ear height and plant aspect suggesting that these traits are under the influence of genetic control. Similarly, Kumar et al. (2014) and Nzuve et al. (2014) also reported the highest GCV and PCV for plant height, ear height, 1000 grain weight and kernel number per row. Hence, these traits can be relied upon; and simple and effective selection can be practiced for further improvement. The inbred lines showed adequate variability with regard to these traits, thus; genetic improvement could be achieved through selection for these traits (Vashistha et al., 2013; Mustafa et

et al., 2015). Low PCV and GCV were observed for leaf width, kernel row per ears, leaf length, days to 50% anthesis, days to 50% silking, and days to maturity and this could probably be attributed to the phenotypic plasticity and also presence of both positive and negative alleles in the maize genotypes leading to low genotypic variation. Similar results were reported by Manju et al. (2002) and Shakoor et al. (2007).

Phenotypic coefficient of variation was found to be higher than the genotypic coefficient of variation for all traits. Similarly, Yusuf (2010) also reported higher phenotypic coefficients of variations than the genotype coefficient of variations for all studied traits. However in this study, the two values differ slightly, indicating less influence of environmental factor on gene expression for the traits. Moreover, the difference between PCV and GCV was low for traits like thousand kernel weight, days to maturity, days to 50% silking, days to 50% anthesis, leaf length, leaf width, plant height, leaf area, leaf length and kernel rows per ears. This implies, less environmental influence on these traits, which ensured practically higher chance for selection. Ear height, number of kernels per row and grain yield showed moderate values between PCV and GCV, which guarantees average chance for selection. Anthesis and silking interval, ear diameter and plant aspect relatively achieved high difference among phenotypic and genotypic coefficient of variation and hence these traits provide practically, less chance for selection, due to higher influence of environmental factors.

Heritability and genetic advance

Broad sense heritability (H^2), an estimate of the total contribution of genetic variance to the total phenotypic variance, ranged from 24.44 for anthesis-silking interval to 96.02 for 1000-kernel weight (Table 3). Higher heritability estimates were scored for 1000-kernel weight (96.02), leaf length (74.79), plant height (69.47), days to 50% anthesis (69.46), days to 50% silking (68.75), leaf width (64.70), ear length (64.62) and leaf area (63.95). Moderate heritability estimates were observed for grain yield per hectare (58.42), ear height (52.99), days to maturity (50.07), number of kernels per row (47.38), plant aspect (33.38) and kernel row per ears (32.26). In contrast, ear diameter (29.82), anthesis and silking interval (24.45) had low heritability estimates.

The high heritability estimates suggest selection of such character could be fairly easy. Therefore, 1000-kernel weight, leaf length, plant height, days to 50% anthesis, days to 50% silking, leaf width and leaf area could easily be passed from one generation to the next then enhancing the efficiency of selection in maize improvement program. This indicated that the traits are under genetic control and the environmental factors did not greatly affect their phenotypic variation. Thus, conventional breeding for these traits could lead to maize improvement (Lule et al., 2012).

Moderate heritability estimates were observed for grain yield per hectare, ear height, days to maturity, number of kernels per row, plant aspect and kernel row per ear indicating these traits may respond positively to phenotypic selection. This is reliable with the result of Al-Tabbal and Al-Fraihat (2012) and Nzuve et al. (2014). The traits exhibited moderate heritability estimates could be improved through heterosis breeding or hybridization (Bello et al., 2012). Ear diameter, anthesis and silking interval indicated low heritability estimates. This implies selection is considerable difficult for such traits due to the masking effect of the environment on the phenotypic traits. The genetic advance as percent of mean (GAM) at 5% selection intensity ranged from 2.76% for days to maturity to 50.69% for grain yield per hectare (Table 3). There was high genetic advance expressed as a percent of mean for some traits like: grain yield (50.69%), 1000-kernel weight (39.44%), ear height (25.13%), ear length (22.94%) and plant height (22.64%). On the other hand, traits such as number of kernels per row (20.00%), ear diameter (19.79%), plant aspect (19.24%), leaf area (19.01%), anthesis-silking interval (16.72%), leaf width (15.67%) and leaf length (13.21%) had moderate genetic advance as percent of mean and the traits like days to 50% anthesis (7.47 %), number of kernel rows per ears (6.90%), days to 50% silking (6.86%) and days to 90% maturity (2.76%) had low genetic advance as percent of mean.

Genetic advance (GA) as a percentage of the mean was higher for traits such as grain yield per hectare, 1000-seed weight and ear height showing that, these traits are under the control of additive gene action. This is supported by the findings of Atnafu and Rao (2014) who reported high genetic advance for plant height, kernel rows per ears, 1000 kernel weight, ear height, and grain yield per hectare. The traits like days to maturity and days to 50% silking indicated low values of genetic advance as per cent of mean and which correspondingly indicated low value of genetic variation for the traits as indicated by low GCV and PCV values. This implies the importance of genetic variability in improvement through selection. This result is also confirmed by results of Fekadu (2014). Maruthi and Rani (2015) observed high genetic advance as per cent of mean for ear height, plant height, number of kernels per ears, ear length, ear diameter and 1000 -grain weight, which is generally in agreement with the result of the present study.

The estimate of GAM for grain yield was 50.69%. The current yield for inbred line CML359 was 5521 kg/ha. Therefore, whenever the best 5% high yielding inbred lines is selected as a parent, the mean grain yield could be improved by 2798.61 kg/ha. As a result, mean genotypic value of the new population of grain yield will be improved from 5521 to 8319.61 kg/ha per one selection cycle for the line CML359. In the same way, the

estimate of genetic advance as percent of mean for 1000-kernel weight was 39.44% and CML359 lines indicated 0.41 kg/ha of 1000-kernel weight. Therefore, after one selection cycle performed for best performing inbred lines at 5% selection intensity, it will be advanced from 0.41 to 0.57 kg/ha. The present study revealed high heritability estimates coupled with the high expected genetic advance as per cent of mean for 1000-kernel weight, ear length and plant height and also moderate heritability estimates with higher genetic advance for grain yield and ear height. This indicated these traits could be improved more easily than the other traits through simple selection. Therefore, even if heritability estimates provide the basis for selection on phenotypic performance, the heritability estimates and genetic advance should be always considered simultaneously, as high heritability is not always associated with high genetic advance.

Conclusion

In this study, considerable amount of genetic variability among the studied inbred lines was observed. The maximum and minimum grain yield per hectare ranged from 5.22t to 0.57t for CML359 and CML159 inbred lines, respectively. PCV was found to be higher than the GCV for all traits. The highest phenotypic and genotypic coefficients of variation were observed for grain yield, anthesis-silking interval, ear diameter, plant aspect and ear height. Leaf length, days to 50% anthesis, days to 50% silking, number of kernel rows per ear and days to maturity had low phenotypic and genotypic coefficient of variation.

High broad sense heritability was recorded for 1000-kernel weight, leaf length, plant height, days to 50% anthesis, days to 50% silking, leaf width, ear length and leaf area which indicated that the variation observed was under genetic control and less influenced by environment. High genetic advance as a percent of mean was observed for grain yield per hectare, 1000-kernel weight, ear height, ears length and plant height. Inbred lines showed high heritability with high genetic advance for thousand kernel weight, ear length and plant height which implies that these traits are under additive gene action. These estimates suggested that selection on the basis of these traits is helpful for breeding program otherwise no genetic gain can be achieved.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Abate T, Bekele S, Abebe M, Dagne W, Yilma K, Kindie T, Menale K, Gezahegn B, Berhanu T, Tolera K (2015). Factors that transformed maize productivity in Ethiopia. Food Security 7(5):965-981.

Aman J, Bantte K, Alamerew S, Tolera B (2015). Evaluation of Quality Protein Maize (Zea mays L) Hybrids at Jimma, Western-Ethiopia. Journal of Forensic Anthropology 1:101.

Atnafu B, Rao TN (2014). Estimates of Heritability, genetic advance and correlation study for yield and its attributes in maize (*Zea mays L.).* Journal of Plant Science 2(1):1-4.

Al-Tabbal JA, Al-Fraihat AH (2012). Heritability studies of yield and yield associated traits in wheat genotypes. Journal of Agricultural Science 4(4):11.

Bello OB, Ige SA, Azeez MA, Afolabi MS, Abdulmaliq SY, Mahamood J (2012). Heritability and Genetic Advance for Kernel Yield and its Component Traits in Maize (*Zea Mays* L.). International Journal of Plant Research 2(5):138-145.

Dudly JW, Moll RH (1969). Interpretation and use of estimates of heritability and genetic variance in plant breeding. Crop Science 9: 257-267.

Falconer DS, Mackay FC (1996). Introduction to Quantitative Genetics. Longman, New York P 464.

Food and Agriculture Organization (FAO) (2013). FAOSTAT Database on Agriculture. Food and Agriculture Organization of United Nations. http://faostat3.fao.org/.

Fekadu K (2014). Genetic variability for yield and yield related traits in some maize (*Zea mays* L.) inbred lines in Central Highland of Ethiopia. Ms.c thesis submitted to the collage of Natural and Computational Sciences, Department of Biology, School of Graduate Studies Haramaya University.

Hallauer AR, Miranda JB (1988). Quantitative genetics in maize breeding. 2nd ed. Iowa state Universality Press.

International board for Plant Genetic Resources (IBPGR) (1991). Descriptors for maize. International maize and wheat Improvement centre, Mexico City/International board for Plant Genetic Resources, Rome. pp. 29-55.

Iqbal J, Shinwari ZK, Rabbani MA (2015). Maize *(Zea mays L.)* Germplasm AAGRO Morphological Characterization Based on Descriptive, Cluster and Principal Component Analysis. Pakistan Journal of Botany 47:255-264.

JARC Agro-metrology department (2016). Mean of ten years (2007-2016) Metrological data of Jimma agricultural research center unpublished.

Johnson HW, Robinson H, Comstock RE (1955). Estimates of genetic and environmental variability in soybeans. Agronomy Journal 47:314-318.

Kebede M, Gezahegne B, Benti T, Mossisa W, Yigzaw D, Assefa A (1993). Maize production trends and research in Ethiopia. Proceedings of the First National Maize Workshop of Ethiopia. Addis Ababa, Ethiopia pp. 142-154.

Kidist A, Nigussie D, Habtamu Z (2013). Growth, Productivity and Nitrogen Use Efficiency of Maize (*Zea Mays* L.) as Influenced by Rate and Time of nitrogen Fertilizer Application in Haramaya District, Eastern Ethiopia. Ms.c thesis.

Kumar GP, Reddy VN, Kumar SS, Rao PV (2014). Genetic Variability, heritability and genetic advance studies in newly developed maize genotypes *(Zea mays L.)*. International Journal of Pure and Applied Bioscience 2(1):272-275.

Lule D, Tesfaye K, Fetene M, De Villiers S (2012). Inheritance and association of quantitative traits in finger millet (Eleusine coracana Subsp. Coracana) landraces collected from eastern and south eastern Africa. International Journal of Genetics 2(2):12-21.

Manju PR, Sreelathakumary I (2002). Genetic variability, heritability and genetic advance in hot chilli (capsicum Chinese jacq.). Journal of Tropical Agriculture. 40:4-6. Retrieved 23 June, 2014 http://jtropag.in/index.php/ojs/article/viewFile/75/70.

Maruthi RT, Rani KJ (2015). Genetic variability, heritability and genetic

advance estimates in maze *(Zea mays L.)* inbred lines. Journal of Applied and Natural Science 7(1):149-154.
Mosisa W, Hadji T, Wonde A, Legese WA, Diallo Twumasi A, A. Guta A (2002). Developing low-N tolerant maize varieties for mid-altitude subhumid agro-ecology of Ethiopia. Integrated Approaches to Higher Maize Productivity in the New Millennium (No. CIS-4176. CIMMYT.).
Mustafa HSB, Farooq J, Bibi T, Mahmood T (2015). Cluster and principle component analyses of maize accessions under normal and water stress conditions. Journal of Agricultural Sciences, Belgrade 60(1):33-48.
Nzuve F, Githri S, Mukunya DM, Gethi J (2014). Genetic variability and Correlation studies of grain yield and related agronomic traits in maize. Journal of Agricultural Science 6(9):166.
Nzuve F, Githiri S, Mukunya DM, Gethi J (2013). Analysis of genotype x environment interaction for grain yield in maize hybrids. Journal of Agricultural Science 5(11):75.
Patterson H, Williams E (1976). A new class of resolvable incomplete block designs. Biometrika 63(1):83-92.
Shakoor MS, Akbar M, Hussain A (2007). Correlation and path coefficient studies of some morpho-physiological traits in maize double crosses. Pakistan Journal Agricultural Science 4(4):213-216. Retrieved from http://pakjas.com.pk/papers%5C315.pdf
Sivasubramanian S, Menon M (1973). Heterosis and inbreeding depression in rice. Madras Agriculture Journal 60:1139.
Sleper DA, Poehlman JM (2006). Breeding field crops (No. Ed. 5). Blackwell publishing.
Taye A (2014). Genetic variability of yield and yield related traits in some maize inbred lines (*Zea mays L.*) developed for mid-altitude agro-ecology of Ethiopia. MSc thesis submitted to the collage of Natural and Computational Sciences, Department of Biology, School of Graduate Studies, Haramaya University.
Tulu B (2014). Correlation and path coefficients analysis studies among yield and yield related traits of quality protein maize (QPM) inbred lines. International Journal of Plant Breeding Crop Science 1(2):6-17.
Vashistha A, Dixit NN, Dipika, Sharma SK, Marker S (2013). Studies on heritability and genetic advance estimates in maize genotypes. Bioscience Discovery 4(2):165-168. Retrieved from http://biosciencediscovery.com 165 ISSN: 2231-024X.
Yusuf M (2010). Genetic variability and correlation in single cross hybrids of quality protein maize (Z. *mays* L.). African Journal of food, Agriculture, nutrition and development 10(2):2166-2175.

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Correlation and path coefficient analysis of yield and quality components of garden cress (*Lepidium sativum* L.) genotypes in Ethiopia

Legesse Tadesse[1,2*], Firew Mekbib[2], Adugna Wakjira[3] and Zerihun Tadele[4]

[1]College of Natural and Computational Sciences, Department of Biology, Dire Dawa University, P.O. Box 1362, Dire Dawa University, Dire Dawa Ethiopia.
[2]College of Agriculture and Environmental Sciences, School of Plant Sciences, Haramaya University, P.O. Box 138, Dire Dawa, Ethiopia.
[3]Ethiopian Institute of Agricultural Research, P. O. Box 2003, Addis Ababa, Ethiopia.
[4]Institute of Plant Sciences, University of Bern, Altenbergrain 21, 3013 Bern, Switzerland.

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The improvement for a trait of interest can be achieved by both direct and indirect selection of characters that are more heritable and easy to select. The aim of this study was to determine the degree and nature of associations among seed yield and seed quality related characters. One hundred eight garden cress (*Lepidium sativum* L.) genotypes were evaluated for their yield and seed quality related traits using a Randomized Complete Block Design with two replications at Raare of Haramaya University Research Site (HRS) and Kulumsa farmer field (KFF), Ethiopia during Meher season in 2014/2015. Correlation and path coefficient analysis were carried out to study the character association and contribution, respectively, for fourteen agro-morphological and seed quality traits. Character association analysis among yield and yield contributing characters revealed that most genotypic correlation coefficients were higher than the respective phenotypic correlation coefficients. Both phenotypic and genotypic correlations revealed that the majority of examined traits had highly significant positive correlation except for oil content and oleoresin content. Genotypic path coefficient analysis of harvest index, biomass per plant and grain yield per plant had exerted positive direct effect on grain yield per plot. Hence, the improvement in grain yield is efficient, if the selection is based on biomass per plant, grain yield per plant and harvest index at both locations.

Key words: Genotypic correlation, oil content, oleoresin content, phenotypic correlation, selection.

INTRODUCTION

Garden cress (*Lepidium sativum* L.), belongs to Brassicaceae Family, is a fast growing annual plant which is cultivated in the temperate and subtropical cold areas throughout the world for its food and medicine

*Corresponding author. E-mail: legese2004@gmail.com.

(Gokavi et al., 2004). It is also an aromatic plant which contains considerable amount of essential oil (Nigist and Sebsebe, 2009) and edible fatty oil rich in its medicinal properties. In addition to its medicinal value, garden cress is used in the form of vegetable (sprout) in North Africa, West and Central Asia and United Kingdom. The breeding program of garden cress relies on improvement of grain yield, oil content of the seeds, resistance to frost, disease and pests (Temesgen et al., 2013a,b; Sabaghnia et al., 2015). The productivity of garden cress is very low; only 600 to 700 kg/ha in India (Sabaghnia et al., 2015) and 1691 to 3415 kg/ha in Ethiopia (Temesgen et al., 2013a). Hence, it requires the attention of plant breeders to evaluate different genotypes across diverse agro-ecological regions in order to obtain high yielding genotypes. Grain yield is the result of a number of complex morphological and physiological processes that interact with each other and with the environment at different growing stages (Semahegn, 2011). The improvement of landraces for grain yield is not only dependent on the nature and extent of genetic variability, heritability and genetic advance in the base population but also on the association of yield and yield-related traits with desirable biochemical composition (Said, 2012; Temesgen et al., 2013a; Sabaghnia et al., 2015). Since sufficient variability is available in the Ethiopian garden cress (Said, 2012; Temesgen et al., 2013a,b), initiating a robust breeding program with this economically important but under-utilized crop will substantially contribute to its genetic improvement, cultivation and utilization.

Genetic evaluation of large number of garden cress genotypes related to yield and seed quality related traits such as oil and oleoresin contents is of paramount importance. Measurement of simple correlation coefficient helps to identify the relative contribution of component characters towards yield (Panse, 1957). Simple correlation coefficients are not always effective in determining the real relationships among traits. Path coefficients show direct influence of independent variable upon dependent variable. It specifies the cause and effect relationship and measures the relative importance of each variable; therefore, path coefficient would provide a more meaningful interpretation of such association (Dewey and Lu, 1959; Mondal et al., 2011; Malek et al., 2014). It is, therefore, worthwhile to consider a large number of garden cress genotypes to make observations at several locations

So far, the study of interrelationships among yield and yield related components characterized with small number of accessions and/or in non-ideal environment of single location (Temesgen et al., 2013a; Said, 2012) make characterization inefficient demanding a large number of genotypes tested in two suitable apparent locations enable the genotypes to discharge its full potential. Therefore, the present study was undertaken to determine the degree and nature of associations among seed yield and seed quality related characters.

MATERIALS AND METHODS

Experimental procedure and data collection

The field experiment was carried out at two locations, namely Haramaya University Agricultural Research Site at Raare (HUARS) and Kulumsa farmer field (KFF) which is 3.4 km East of Kulumsa Agricultural Research Center, Ethiopia, in 2014/2015 growing season. One hundred eight garden cress accessions collected from different agro-ecological regions of Ethiopia were examined. The experiment was conducted in the Randomized Complete Block Design with two replications. Each experimental plot consisted of two 135 cm long rows with inter-row and between plant spacing of 20 and 15 cm, respectively. Fertilizers were applied at the rate of 18 kg/ha N and 46 kg/ha P_2O_5 and all necessary cultural practices were undertaken as recommended for the crop. On plant basis, data were recorded from five randomly selected competitive plants of each genotype characters namely plant height (PH), number of primary branches per plant (PB), number of secondary branches per plant (SB), grain yield per plant (GYPP), and biomass yield per plant (BYPP). On plot basis, traits such as days to flowering initiation (FI), days to 50% flowering (FL), days to maturity (DM), thousand seed weight (TSW), grain yield per plot (GYPlt), harvest index (HI), and biomass yield per plot (BMPlt) were recorded. The agronomic characters were taken after harvesting the plants. The oil content of the seed was determined at Holetta Highland Oil Crops Laboratory in Ethiopia (Guenther, 2007) while the oleoresin content was investigated at Wondo Genet Natural Product Laboratory, Ethiopia (Daniel et al., 2008).

Data analysis

Correlation coefficients

The correlation coefficients were calculated to determine the degree of association of characters with yield and among themselves. Phenotypic and genotypic correlations were computed by using the standard procedure suggested by Johnson et al. (1955), Singh and Chaudhary (1985) and Dabholkar (1992) using the following formula:

$$r_{gxy} = [Cov_{gxy}/(\sqrt{(\sigma^2_{gx} x \sigma^2_{gy})}]$$

$$r_{pxy}=[Cov_{pxy}/(\sqrt{(\sigma^2_{px} x \sigma^2_{py})}].$$

where r_{pxy} = phenotypic correlation coefficient between characters x and y, Cov_{pxy} = phenotypic covariance between characters x and y, σ^2_{px} = phenotypic variance for character x and σ^2_{py} = phenotypic variance for character y, r_{gxy} = genotypic correlation coefficient between characters x and y, Cov_{gxy} = genotypic covariance between characters x and y, σ^2_{gx}= genotypic variance for character x and σ^2_{gy} = genotypic variance for character y. The coefficients of correlation were tested using 'r' tabulated value at n - 2 degrees of freedom, at 5 and 1% probability levels, where n is the number of treatments (genotypes) as described by Robertson (1959).

Path analysis

Path analysis was used for exhibiting the direct and indirect effects on seed yield according to the method suggested by Dewey and Lu (1959) using phenotypic as well as genotypic correlation coefficients as:

$$rij = pij + \Sigma rikpkj$$

where rij = mutual association between the independent character i (yield-related trait) and dependent character, j (grain yield) as measured by the genotypic correlation coefficients, Pij = components of direct effects of the independent character (i) on the dependent character (j) as measured by the path coefficients, and ∑*rikpkj* = summation of components of indirect effects of a given independent character (i) on a given dependent character (j) *via* all other independent characters (k), where: I = any trait in the model, and r = correlation coefficient between any trait i and the dependent variable.

Data processing was done by MS Excel 2007. The contribution of the remaining unknown characters was measured as the residual effect as demonstrated by Singh and Chaudhary (1985). SPAR2.0 (IASRI, 2005) and META-R version 5 (Alvarado et al., 2015) statistical packages were used for analysis.

RESULTS AND DISCUSSION

Phenotypic and genotypic correlations

Estimates of genotypic and phenotypic correlation coefficients at Haramaya and Kulumsa environments are shown in Tables 1 and 2, respectively. The analysis of correlations in each location showed that genotypic correlation coefficients were generally higher than their corresponding phenotypic correlations indicating strong hereditary association among the traits due to genetic factors such as linkage and/or pleiotropic effect enabling consistent performance across wide range of environments as described by Waitt and Levin (1998). Similar findings were observed by Tahira et al. (2011) and Hasan et al. (2015) on the study of *Brassica*. Grain yield is a very complex character which is governed by polygenes and the result of numerous simple characters presumably showing considerable variations from one environment to another (Singh et al., 2014). The correlation between the traits may be due to linkage or pleiotropy (Allard, 1999) or environment (Aastveit and Aastveit, 1993). Similarly, a narrow difference between phenotypic and genotypic correlation coefficient was obtained for almost all the pairs of characters studied showing that masking or modifying effects of the environment was little demonstrating that the presence of an inherent association among these characters. Similar results were reported in barley (Azeb et al., 2016).

Correlations of grain yield with other traits

It was observed that majority of the phenotypic and genotypic correlation coefficients between examined traits were highly significant ($p < 0.001$) at both environments. Grain yield per plot showed highly and positively significant correlations with days to flowering initiation, days to 50% flowering, plant height, number of primary branches, number of secondary branches and biomass per plant. It was also positively and significantly correlated ($p < 0.001$) with biomass per plot, grain yield per plant and thousand seed weight at both environments. However, phenotypic correlation was not significant for harvest index due to higher biomass per plant at Haramaya and Kulumsa environments, respectively (Tables 1 and 2). Similar findings were reported for rapeseed where significantly positive association were found between grain yield per plot and days to flowering initiation and days to 50% flowering and number of primary branches (Gangapur et al., 2009; Temesgen et al., 2013a; Tesfaye et al., 2013; Halder et al., 2016).

At HRS, the highest and significant genotypic and phenotypic correlations between grain yield per plot and grain yield per plant were 0.980 and 0.967, respectively. Similarly, at KFF, positive and highly significant genotypic and phenotypic correlations were recorded for the same traits with the respective values of 0.981 and 0.979. In addition, genotypic and phenotypic correlations between grain yield and the following traits were positive and significant, respectively for number of primary branches (0.727 and 0.707), number of secondary branches (0.689 and 0.685), biomass per plot (0.689 and 0.688), and biomass per plant (0.875 and 0.854) at KFF (Table 2). Hasan et al. (2015) also reported that plant height, number of primary branches and grain yield per plant had positive and strong correlation with grain yield per plot in *Brassica juncea*. Along with these, at KFF, biomass per plant again showed positive and significant genotypic and phenotypic correlations, respectively with number of primary branches (0.776 and 0.753), number of secondary branches (0.736 and 0.729), and grain yield per plants (0.844 and 0.820) (Table 2). Similarly, in Indian mustard, significant genetic and phenotypic correlations were obtained between grain yield and grain yield per plant, number of primary and secondary branches per plant, biomass per plant, and thousand seed weight (Gangapur et al., 2009).

On the contrary, grain yield per plot exhibited a non-significant with oleoresin content with oil content. The findings were similar for both genotypic and phenotypic correlations at both environments. In contrary, Temesgen et al. (2013a) reported that oil content had significant positive correlation to grain yield probably due to the influence of the environment. Non-significant values of correlation coefficient were observed between oleoresin contents and most of the studied characters in both environments except for thousand seed weight and oil content indicating independence of traits in both environments.

Correlation among yield contributing traits

Significant genotypic and phenotypic correlations were observed respectively between days to maturity and days to 50% flowering (0.883 and 0.873), plant height and days to maturity (0.865 and 0.855), and biomass per plant and biomass per plot (0.837, 0.821) at HRS. Significantly, high and positive genotypic and phenotypic

Table 1. Genotypic (below diagonal) and phenotypic (above diagonal) correlation coefficient between traits of 108 garden cress (*Lepidium sativum* L.) genotypes at Haramaya Research Site (HRS) at Raare, Ethiopia in 2014/2015 growing season.

Trait	FI	FL	PH	PB	SB	DM	BMPP	BMPlt	GYPP	HI	TSW	OC	OR	GYPLT
FI	1	0.933**	0.812**	0.574**	0.650**	0.867**	0.506**	0.409 **	0.369**	-0.077ns	0.033ns	-0.172^{ns}	0.132^{ns}	0.380**
FL	0.937**	1	0.792**	0.551**	0.618**	0.873**	0.510**	0.413 **	0.365**	-0.081ns	0.02ns	-0.201*	0.109^{ns}	0.375**
PH	0.820**	0.803**	1	0.642**	0.751**	0.855**	0.513**	0.566 **	0.494**	0.056ns	0.143ns	-0.122^{ns}	0.094^{ns}	0.520**
PB	0.599**	0.576**	0.674**	1	0.765**	0.546**	0.453**	0.499**	0.515**	0.160ns	0.142ns	-0.105^{ns}	0.048^{ns}	0.536**
SB	0.656**	0.623**	0.766**	0.804**	1	0.672**	0.495**	0.548 **	0.588**	0.174ns	0.246**	-0.091^{ns}	0.148^{ns}	0.573**
DM	0.875**	0.883**	0.865**	0.572**	0.680**	1	0.558**	0.489 **	0.473**	-0.008ns	0.094ns	-0.169^{ns}	0.161^{ns}	0.478**
BMPP	0.522**	0.528**	0.532**	0.476**	0.507**	0.577**	1	0.821 **	0.632**	-0.300**	0.464**	-0.106^{ns}	0.100^{ns}	0.635**
BMPlt	0.421**	0.425 **	0.567 **	0.514 **	0.548**	0.499 **	0.837 **	1	0.664**	-0.068ns	0.422**	-0.099^{ns}	0.103^{ns}	0.671**
GYPP	0.379**	0.378**	0.511**	0.538**	0.597**	0.485**	0.659**	0.673**	1	0.511**	0.544**	-0.142^{ns}	0.129^{ns}	0.967**
HI	-0.087^{ns}	-0.087	0.058^{ns}	0.181^{ns}	0.185^{ns}	-0.015^{ns}	-0.267**	-0.077^{ns}	0.531**	1	0.191*	-0.060^{ns}	0.049^{ns}	0.526**
TSW	0.034^{ns}	0.022	0.147^{ns}	0.166^{ns}	0.264**	0.101^{ns}	0.496**	0.435**	0.585**	0.194*	1	-0.059^{ns}	-0.116^{ns}	0.598**
OC	-0.170^{ns}	-0.200*	-0.123^{ns}	-0.112^{ns}	-0.091^{ns}	-0.169^{ns}	-0.109^{ns}	-0.097^{ns}	-0.146^{ns}	-0.063^{ns}	-0.062^{ns}	1	-0.035^{ns}	-0.161^{ns}
OR	0.156^{ns}	0.128^{ns}	0.116^{ns}	0.045^{ns}	0.166^{ns}	0.184^{ns}	0.124^{ns}	0.123^{ns}	0.146^{ns}	0.041^{ns}	-0.147^{ns}	-0.038^{ns}	1	0.104^{ns}
GYPLT	0.382**	0.380**	0.528**	0.559**	0.577**	0.481**	0.653**	0.666**	0.980**	0.539**	0.623**	-0.163^{ns}	0.112^{ns}	1

*,** and ns: significant at $p < 0.05$ (0.190), $p < 0.01$ (0.241) and non-significant ($p > 0.05$), probability levels, respectively; FI: Days to flowering initiation, FL: days to 50% flowering, PH: plant height, PB: number of primary branches, SB: number of secondary branches, DM: days to maturity, BMPP: biomass per plant , BMPlt: biomass per plot, GYPP: grain yield per plant, HI: harvest index; TSW: 1000-seed weight, GYPLT: seed yield per plot, OC: oil content, OR: oleoresin content.

correlations were obtained between number of primary branches and number of secondary branches (0.804 and 0.765, respectively) (Table 1). Similarly, at KFF, highly significant genotypic and phenotypic correlations were recorded, respectively between flowering initiation and days to 50% flowering (0.907 and 0.894), grain yield per plant and biomass per plant (0.844 and 0.820), days to 50% flowering and days to maturity (0.879 and 0.859), days to maturity and days to flowering initiation (0.841 and 0.831) and plant height and days to 50% flowering (0.805 and 0.772). Similar findings were reported by Temesgen et al. (2013a) and Islam and Haque (2015). These results are in agreement with the reports of Hasan et al. (2015) where plant height significantly and positively correlated with thousand seed weight, grain yield per plant and grain yield per plot while thousand seed weight was negatively correlated with grain yield per plant at genotypic level. They also reported that days to flowering has strong and significant positive phenotypic and genotypic correlation with days to maturity and number of primary branches in *B. juncea.*

It was also observed that oil content showed significant and negative correlation with flowering initiation, days to 50% flowering at KFF unlike at HRS. Even though, oil and oleoresin contents showed non-significant correlation with most traits at both locations (Tables 1 and 2). These results are in agreement with an earlier study (Beemnet et al., 2013) which reported on coriander accessions that essential oil and oil contents were non-significantly correlated with the majority of yield and yield related components such as plant height, thousand seed weight, days to maturity and grain yield per plant. Tesfaye et al. (2013) also reported days to flowering, days to maturity, plant height, number of primary and secondary branches biomass per plot non-significantly correlated with harvest index in their study of *Brassica carinata*. On the other hand, Temesgen et al. (2013a) in their study of 49 Ethiopian garden cress genotypes reported that majority of the examined traits showed significant and positive correlation with oil content. The negative correlation indicated that it would not be possible to improve both traits simultaneously depending on the linkage intensity or the degree of compromise between the two traits (Dabholkar, 1992). The oil and oleoresin contents will be disregarded in selection for grain yield improvement programs (Ariyo et al. 1987; Henry

Table 2. Genotypic (below diagonal) and phenotypic (above diagonal) correlation coefficient between traits of 108 garden cress (*Lepidium sativum* L.) genotypes at Kulumsa farmer field (KFF) at 3.4 km East of Kulumsa Agricultural Research Center, Ethiopia in 2014/15 growing season.

Trait	FI	FL	PH	PB	SB	DM	BMPP	BMPPlt	GYPP	HI	TSW	OC	OR	GYPLT
FI	1	0.894**	0.774**	0.533**	0.658**	0.831**	0.377**	0.433**	0.296**	-0.205**	-0.261**	-0.202*	0.169^{ns}	0.306**
FL	0.907**	1	0.772**	0.557**	0.679**	0.859**	0.414**	0.474**	0.341**	-0.233**	-0.249**	-0.184^{ns}	0.177^{ns}	0.340**
PH	0.797**	0.805**	1	0.624**	0.685**	0.754**	0.489**	0.609**	0.430**	-0.164^{ns}	-0.154^{ns}	-0.173^{ns}	0.103^{ns}	0.448**
PB	0.562**	0.581**	0.653**	1	0.856**	0.494**	0.753**	0.625**	0.687**	-0.188^{ns}	0.135^{ns}	-0.048^{ns}	0.030^{ns}	0.707**
SB	0.677**	0.704**	0.711**	0.877**	1	0.626**	0.729**	0.612**	0.676**	-0.194^{ns}	0.055^{ns}	-0.028^{ns}	0.050^{ns}	0.685**
DM	0.841**	0.879**	0.771**	0.520**	0.642**	1	0.374**	0.486**	0.331**	-0.140^{ns}	-0.257**	-0.135^{ns}	0.139^{ns}	0.341**
BMPP	0.387**	0.433**	0.500**	0.776**	0.736**	0.383**	1	0.730**	0.820**	-0.400**	0.350**	-0.123^{ns}	0.084^{ns}	0.854**
BMPlt	0.436**	0.483**	0.621**	0.653**	0.618**	0.490**	0.750**	1	0.686**	-0.221	0.108^{ns}	-0.186^{ns}	0.025^{ns}	0.688**
GYPP	0.292**	0.343**	0.430**	0.707**	0.677**	0.332**	0.844**	0.686**	1	0.142^{ns}	0.368**	-0.029^{ns}	0.007^{ns}	0.979**
HI	-0.232*	-0.275**	-0.170^{ns}	-0.179^{ns}	-0.180^{ns}	-0.148^{ns}	-0.347**	-0.240*	0.164^{ns}	1	0.017^{ns}	0.135^{ns}	-0.102^{ns}	0.109^{ns}
TSW	-0.275**	-0.275**	-0.180^{ns}	0.131^{ns}	0.051^{ns}	-0.284**	0.358**	0.119^{ns}	0.403**	0.078^{ns}	1	0.101^{ns}	-0.104^{ns}	0.378**
OC	-0.201*	-0.186^{ns}	-0.176^{ns}	-0.049^{ns}	-0.028^{ns}	-0.136^{ns}	-0.127^{ns}	-0.186^{ns}	-0.030^{ns}	0.151^{ns}	0.112^{ns}	1	-0.035^{ns}	-0.058^{ns}
OR	0.191^{ns}	0.200*	0.137^{ns}	0.055^{ns}	0.086^{ns}	0.172^{ns}	0.115^{ns}	0.030^{ns}	0.010^{ns}	-0.161^{ns}	-0.138^{ns}	-0.038^{ns}	1	0.018^{ns}
GYPLT	0.304**	0.342**	0.452**	0.727**	0.689**	0.343**	0.875**	0.689**	0.981**	0.203*	0.409**	-0.059^{ns}	0.024^{ns}	1

*,** and ns: significant at $p < 0.05$ (0.190), $p < 0.01$ (0.241) and non-significant ($p > 0.05$), probability levels, respectively; FI: Days to flowering initiation, FL: days to 50% flowering, PH: plant height, PB: number of primary branches, SB: number of secondary branches, DM: days to maturity, BMPP: biomass per plant , BMPlt: biomass per plot, GYPP: grain yield per plant, HI: harvest index; TSW: 1000-seed weight, GYPLT: seed yield per plot, OC: oil content, OR: oleoresin content.

and Krishna, 1990).At both locations, five agro-morphological traits (namely, grain yield per plant, shoot biomass per plant, shoot biomass per plot, number primary branches per plant and number of secondary branches per plant) are significantly correlated among themselves. Hence, selection in the performance of one of the traits will result in the improvement of the other traits. However, such traits need further study on their direct and indirect effect on the grain yield per plot by path coefficient analysis to remark as selection criteria for grain yield improvement programs.

Genotypic path analysis

In the present study, grain yield per plot (GYPLt) was the dependent variable while other evaluated traits were considered as independent variables. Direct and indirect effects of these components were determined on grain yield and their contributions in each of the two environments are shown in Table 3. The path coefficient analysis at genotypic level revealed at HRS and KFF, respectively that biomass per plant (1.143 and 0.996) had the highest positive direct effect on yield followed by harvest index (0.694 and 0.274) and grain yield per plant (0.171 and 0.134). Similarly, harvest index and grain yield per plant had highly significant and positive correlations with grain yield in each location at genotypic level, indicating the need for direct selection for these traits in order to improve grain yield of garden cress. At HRS, the highly significant direct correlation of grain yield with days to flowering initiation (0.126), with plant height (0.274) and with number of secondary branches (0.182) (Table 3) shows the usefulness of selecting of these traits for grain yield improvement. Similar findings were reported by Temesgen et al. (2013a) on their study with the Ethiopian garden cress. However, Hasan et al. (2015) indicated that plant height had negative direct effect while grain yield per plant had strong positive and direct effect on grain yield of *Brassica napus* at genotypic level indicating correlation of traits varied depending on the environment and nature of the crop.

It was observed that plant height, days to maturity and oil content exhibited positive and relatively low direct effect on yield at KFF (Table 3). These traits except for oil content had highly

Table 3. Direct (bold and underlined) and indirect effects (off diagonal) of traits on grain yield at genotypic level of 108 garden cress (*Lepidium sativum* L.) genotypes at Haramaya Research Site (HRS) at Raare and Kulumsa farmer field (KFF) at 3.4 km East of Kulumsa Agricultural Research Center, Ethiopia in 2014/15 growing season.

Loc*	Trait	FI	FL	PH	PB	SB	DM	BMPP	BMPlt	GYPP	HI	TSW	OC	OR	rg
HRS	FI	**0.126**	-0.23	0.232	-0.172	0.123	-0.166	0.642	-0.107	0.07	-0.067	-0.008	0.009	-0.057	0.395**
KFF		**-0.065**	**0**.014	0.064	-0.027	-0.029	0.037	0.428	-0.042	0.04	-0.095	0.012	-0.002	-0.027	0.310**
HRS	FL	0.119	**-0.244**	0.228	-0.165	0.116	-0.168	0.650	-0.106	0.07	-0.063	-0.006	0.01	-0.045	0.397**
KFF		-0.06	**0.015**	0.066	-0.027	-0.031	0.04	0.489	-0.047	0.048	-0.12	0.013	-0.002	-0.028	0.357**
HRS	PH	0.107	-0.203	**0.274**	-0.191	0.145	-0.166	0.657	-0.142	0.094	0.046	-0.034	0.006	-0.048	0.544**
KFF		-0.055	0.013	**0.076**	-0.03	-0.031	0.035	0.540	-0.063	0.059	-0.058	0.01	-0.002	-0.025	0.469**
HRS	PB	0.085	-0.159	0.207	**-0.253**	0.165	-0.121	0.616	-0.139	0.103	0.164	-0.049	0.007	-0.009	0.615**
KFF		-0.041	0.01	0.055	**-0.042**	-0.038	0.025	0.842	-0.068	0.102	-0.043	-0.004	-0.001	-0.015	0.783**
HRS	SB	0.085	-0.156	0.218	-0.229	**0.182**	-0.131	0.614	-0.134	0.106	0.148	-0.066	0.005	-0.057	0.585**
KFF		-0.046	0.011	0.058	-0.039	**-0.041**	0.029	0.770	-0.062	0.094	-0.043	-0.001	0.00	-0.021	0.709**
HRS	DM	0.112	-0.22	0.244	-0.165	0.128	**-0.186**	0.711	-0.125	0.088	-0.014	-0.025	0.009	-0.063	0.494**
KFF		-0.056	0.014	0.061	-0.024	-0.027	**0.043**	0.409	-0.047	0.046	-0.051	0.014	-0.002	-0.029	0.351**
HRS	BMPP	0.071	-0.138	0.157	-0.136	0.098	-0.116	**1.143**	-0.212	0.124	-0.125	-0.124	0.006	-0.052	0.695**
KFF		-0.028	0.007	0.041	-0.035	-0.031	0.018	**0.996**	-0.074	0.120	-0.048	-0.014	-0.002	-0.025	0.926**
HRS	BMPlt	0.057	-0.109	0.164	-0.149	0.103	-0.098	1.023	**-0.237**	0.124	-0.055	-0.1	0.005	-0.041	0.687**
KFF		-0.029	0.008	0.051	-0.031	-0.027	0.022	0.801	**-0.092**	0.092	-0.08	-0.005	-0.002	-0.003	0.704**
HRS	GYPP	0.051	-0.100	0.151	-0.152	0.112	-0.096	0.829	-0.171	**0.171**	0.407	-0.148	0.008	-0.051	0.999**
KFF		-0.02	0.005	0.034	-0.032	-0.029	0.015	0.897	-0.063	**0.134**	0.069	-0.019	0.00	-0.003	0.987**
HRS	HI	-0.012	0.022	0.018	-0.06	0.039	0.004	-0.206	0.019	0.1	**0.694**	-0.044	0.003	-0.004	0.573**
KFF		0.022	-0.007	-0.016	0.007	0.006	-0.008	-0.174	0.027	0.034	**0.274**	-0.012	0.002	0.047	0.203*
HRS	TSW	0.005	-0.006	0.043	-0.058	0.056	-0.022	0.663	-0.110	0.118	0.142	**-0.214**	0.003	0.066	0.685**
KFF		0.022	-0.006	-0.021	-0.005	-0.001	-0.016	0.399	-0.014	0.071	0.091	**-0.036**	0.002	0.03	0.516**
HRS	OC	-0.022	0.049	-0.035	0.032	-0.017	0.032	-0.134	0.024	-0.026	-0.048	0.015	**-0.051**	0.013	-0.167^{ns}
KFF		0.013	-0.003	-0.014	0.002	0.001	-0.006	-0.137	0.018	-0.004	0.058	-0.005	**0.012**	0.005	-0.060^{ns}
HRS	OR	0.033	-0.051	0.062	-0.01	0.048	-0.055	0.276	-0.045	0.041	0.012	0.066	0.003	**-0.215**	0.165^{ns}
KFF		-0.02	0.005	0.022	-0.007	-0.01	0.014	0.278	-0.003	0.005	-0.148	0.012	-0.001	**-0.088**	0.061^{ns}

Residual effect: HRS=9.8%; KFF = 7.2%Loc = location, HRS: Haramaya Research Site at Raare, KFF: Kulumsa farmer field, Var: variable , FI: flowering Initiation, FL: days to 50% flowering, PH: plant height, PB: number of primary branches, SB: number of secondary branches, DM: days to maturity, BMPP: biomass per plant (g), BMPlt: biomass per plot, GYPP: grain yield per plant, HI: harvest index; TSW: 1000-Seed Weight, OC: oil content, OR: oleoresin content, rg: genotypic correlation coefficient of grain yield per plot with other yield related traits.

significant positive correlation with grain yield in an earlier study (Temesgen et al., 2013a). On the contrary, the number of primary branches, biomass per plot, thousand seed weight, and oleoresin content were negative and low direct effect for grain yield in both locations. Days to 50% flowering and days to maturity at HRS and days to flowering initiation and number of secondary branches at KFF had negative and low direct effect to grain yield but positive and significant correlation with grain yield except for oleoresin content which was non-significant

(Table 3). These implied that almost all of these traits might probably contribute for grain yield through indirect effects requiring indirect selection. Halder et al. (2016) demonstrated similar high positive direct effect of plant height, thousand seed weight, and days to 50% flowering on grain yield/ha. These results are also in agreement with Marjanovic Jeromela et al. (2008) and Halder et al. (2016) who reported that plant height exerted the highest positive significant direct effect on grain yield. In the study of rapeseed, Uddin et al. (2013) and Ara et al. (2013) also reported significant direct effect from primary branches per plant and number of secondary branches per plant on grain yield.

Traits such as days to flowering initiation, days to 50% flowering, plant height, number of primary and secondary branches, days to maturity, biomass per plot, grain yield per plant and thousand seed weight exerted indirect effects on grain yield *via* biomass per plant in both environments (Table 3). This demonstrated that the relationship between these traits and grain yield in the two environments were predominantly due to indirect effects. Harvest index showed relatively higher negative indirect effect to grain yield through biomass per plant. According to Temesgen et al. (2013a), all investigated traits showed direct effects to grain yield except for days to 50% flowering and plant height which revealed indirect effect. This was probably due to the type of genotypes used in the study and environmental factors that could also substantially contributed for the deviation of the result.

Generally, the direct and indirect impact of traits in path analysis in each location depicted considerable variation. This was presumably due to significant differential performance across location which resulted in substantial changes in the direction and magnitude of genotypic correlations (Table 3). Accordingly, the grain yield per plot of garden cress can be increased by selecting genotypes having higher biomass per plant, harvest index and grain yield per plant at both environments and higher plant height and number of secondary branches at HRS. This is in agreement with earlier work on lentil by Hegazy et al. (2012). As shown in Table 3, most traits at both locations exerted their indirect effect to increased grain yield through biomass per plant. Similar findings were reported from *B. juncea* (Hasan et al., 2015).

Path coefficient analysis revealed that biomass per plant, harvest index, and grain yield per plant had high positive direct effects on yield at both environments and plant height, flowering initiation and number of secondary branches showed direct and significant contributions to grain yield at HRS unlike at KFF. A higher positive indirect contribution by most of the yield components *via* biomass per plant was responsible for highly significant genotypic and phenotypic correlation coefficients. It could be said that biomass per plant, harvest index, and grain yield per plant were consistent and the most important traits affecting the yield per plot of garden cress. Thus, due attention must be given to these traits in the selection program. Along with these, days to flowering initiation, days to 50% flowering, plant height, number of primary and secondary branches, days to maturity, biomass per plot, and thousand seed weight contributed indirectly to grain yield at both locations. Therefore, including these traits directly or indirectly in the selection criteria is important towards developing high grain yielding garden cress genotypes.

In the present study, residual effect at HRS and KFF were 0.098 and 0.072, respectively (Table 3). This means, characters in the path analysis expressed in grain yield contributing traits by 90.2% at HRS and 93.8% at KFF, while the remaining needs additional characterization for the future breeding program.

Conclusions

The genotypic correlation coefficient was generally higher than the corresponding phenotypic correlation coefficient. Genotypic and phenotypic correlations between grain yield and majority of investigated traits were highly significant at each location except oil and oleoresin contents. Plant height, days to flowering initiation and number of secondary branches had high direct contribution to the grain yield at HRS unlike at KFF. Genotypic path coefficient analysis revealed that biomass per plant, grain yield per plant, and harvest index had relatively higher positive direct effect on grain yield in each location. In general, this study has clearly indicated the need for focusing on biomass per plant, grain yield per plant, and harvest index traits towards improving grain yield of garden cress.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Aastveit AH, Aastveit K (1993). Effects on genotype environment interactions on genetic correlation. Theoretical and Applied Genetics 86:1007-1013.

Allard RW (1999). Principles of Plant Breeding. 2nd edition, John Willey and Sons, Inc., New York.

Alvarado G, Lopez M, Vargas M, Pacheco A, Rondriqrez F, Burgueho J, Cross J (2015). META-R *(*Multi Environment Trial Analysis with R) for windows version 5 CIMMYT, Mexico.

Ara S, Afroz S, Noman MS, Bhuiyan SR, Zia IK (2013). Variability, correlation and path analysis in F2 progenies of inter-varietal crosses of *Brassica rapa.* Journal of Environmental Sciences and Natural Resources 6(1):217-220.

Ariyo OJ, Aken'ova ME, Fatokun CA (1987). Plant character correlation and path analysis of pod yield in okra (*Abelmoschusesculentus)*. Euphytica 36:677-686.

Azeb H, Sentayehu A, Mandefro N, Ermias A (2016). Correlation and path coefficient analysis of yield and yield associated traits in barley (*Hordeum vulgare* L.) germplasm. Advances in Crop Sciences Technology 4(2):1-9.

Beemnet M, Alemaw G, Tesfaye B (2013). Correlation studies and path coefficient analysis for seed yield and yield components in Ethiopian coriander accessions. African Crop Science Journal 21(1):51-59.
Dabholkar AR (1992) Elements of biometrical genetics. Concept Publishing Company, New Delhi, India.
Daniel B, Solomon A, Wossen K (2008) Laboratory manual for plant product analysis. Ethiopia. Technical Manual 1(23):3-10.
Dewey DR, Lu KH (1959). A correlation and path coefficient of components of crested wheat grass seed production. Agronomy Journal 51:515-518.
Gangapur D, Prakash BG, Salimath PM, Ravikumar RL, Rao SL (2009). Correlation and path analysis in Indian mustard (*Brassica juncea* L. Czernand Coss). Karnataka Journal of Agricultural Sciences 22(5):971-977.
Gokavi SS, Malleshi NG, Guo M (2004). Chemical composition of garden cress (*Lepidium sativum* L.) seeds and its fractions and use of bran as a functional ingredient. Plant Foods for Human Nutrition 59:105-111.
Guenther E (2007). The Essential Oils - Vol 1: History, Origin in Plants, Production, Analysis. Jepson Press.
Halder T, Bhuiyan SR, Islam, MS, Hossain J (2016). Analysis of relationship between yield and some yield contributing characters in few advanced lines of rapeseed (*Brassica rapa*) by using correlation and path analysis. International Journal of the Bioflux Society 8(1):36-44.
Hasan EU, Bibi MH, Mahmood T, Tanveer M, Kalyar A, Salim J (2015). Genetic evaluation and characterization for yield and related traits in mustard (*Brassica juncea*). Research Journal of Agriculture and Environmental Management 4(2):082-087.
Hegazy RE, Selim T El-Emam EA (2012). Correlation and path coefficient analyses of yield and some yield components in lentil. Egypt Journal of Plant Breeding 16 (3):147-159.
Henry A, Krishnna GV (1990). Correlation and path coefficient analysis in pigeon pea. Madras African Journal 77(9-12):443-446.
Indian Agricultural Statistics Research Institute (IASRI) (2005). Statistical Package for Agricultural Research (Windows Version) SPAR 2.0. IASRI, Library Avenue, New Delhi
Islam MS, Haque MM (2015). Estimation of genotypic and phenotypic coefficients variation of yield and its contributing characters of *Brassica rapa* L. American-Eurasian Journal of Agriculture and Environmental Sciences 15(10):2029-2034.
Johnson HW, Robinson HF, Comstock RE (1955). Estimation of genetic and environmental variability in soybean. Agronomy Journal 47:314-318.
Malek MA, Rafii Y, Afroz M, Sharmin S, Nath UK, Mondal MMA (2014). Morphological characterization and assessment of genetic variability, character association and divergence in soybean mutants. The Scientific World Journal 2014:1-12.
Marjanovic-Jeromela A, Marinkovic R, Mijic A, Zdunic Z, Ivanovska S, Jankulovska M (2008). Correlation and path analysis of quantitative traits in winter rapeseed (*Brassica napus* L.). Agriculture Conspectus Sciences 73(1):13-18.
Mondal MA, Hakim MA, Juraimi AS, Azad M (2011). Contribution of morpho-physiological attributes in determining yield of mung bean. African Journal of Biotechnology 10:12897-12904.
Nigist A, Sebsebe D (2009). Aromatic Plants of Ethiopia. 1st Ed. Shaama Books, Addis Ababa, Ethiopia.
Panse VG (1957). Genetics of quantitative characters in relation to plant breeding. Indian Journal of Genetics 17:318-328.
Robertson A (1959). The sampling variance of the genetic correlation coefficient. Biometrics 15:469-485.
Sabaghnia N, Ahadnezhad A, Janmohammdi M (2015). Genetic variation in garden cress (*Lepidium sativum* L.) germplasm as assessed by some morphological traits. Genetic Resources and Crop Evolution 62:733-745.
Said M (2012). Genetic diversity study of *Lepidium sativum* L. populations from Ethiopia using morphological characters and ISSR markers. MSc. Thesis, Addis Ababa University.
Semahegn Y (2011). Genetic variability, correlation and path analysis studies in Ethiopian mustard (*Brassica carinata* A. Brun) genotypes. International Journal of Plant Breeding and Genetics 5(4):328-338.
Singh JL, Prasad C, Madakemohekar AH. Bornare SS (2014). Genetic variability and character association in diverse genotypes of barley (*Hordeum vulgare* L.). The Bioscan (Supplement on Genetics and Plant Breeding) 9(2):759-761.
Singh RK, Chaudhary BD (1985). Biometrical methods in quantitative genetic analysis. Revised, Kalyani Publishers, Ludhiana, New Delhi, India.
Tahira T, Mahmood MS, Tahir U, Saleem M, Hussain MS (2011). The estimation of heritability, association and selection criteria for yield components in mustard (*Brassica juncea*). Pakistan Journal of Agricultural Sciences 48(4):251-254.
Temesgen B, Mebeaselassie A, Million E (2013a). Genetic variability and association among yield, yield related traits and oil content in Ethiopian garden cress (*Lepidium sativum* L.) genotypes. Journal of Plant Breeding and Crop Sciences 5:141-149.
Temesgen B, Mebeaselassie A, Million E (2013b). Genetic divergence analysis of garden cress (*Lepidium sativum* L.). Journals of Plant Breeding and Crop Sciences 5:770-774.
Tesfaye W, Adugna W, Tsige G (2013). Correlation and path coefficient analysis among yield component traits (*Brassica Carinata* A. Brun) in Ethiopian mustard at Adet, Northwestern, Ethiopia. International Journal of Cereals and Oilseeds 1(1):01-16.
Uddin MS, Bhuiyan SR, Mahmud F, Kabir K (2013). Study on correlation and path coefficient in F_2 progenies of rapeseed. Academic Journal Plant Sciences 6(1):13-18.
Waitt DE, Levin DA (1998). Genetic and phenotypic correlations in plants: a botanical test of Cheverud's conjecture. Heredity 80:310-319.

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Genetic variability of some chickpea (*Cicer arietinum* L.) genotypes and correlation among yield and related traits in humid tropics of southern Ethiopia

Mieso Keweti Shengu*, Dereje Hirpa and Zenabu Wolde

Department of Plant Science, College of Agriculture and Natural Resources, Dilla University, P.O. Box 419, Dilla, Ethiopia.

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The current investigation evaluated the genetic variability of some chickpea (*Cicer arietinum* L.) genotypes and correlation among yield and related traits in Abaya Woreda. Five improved chickpea genotypes along with one local variety were laid out in randomized complete block design with three replications. Data were recorded from phenological, growth parameters; and yield and related traits depicted the ranges of mean values for most of the traits were large depicting the existence of genetic variations among the tested genotypes. Phenotypic coefficients of variation (PCV) were found to be higher than genotypic coefficients of variation (GCV) for all the traits. Higher heritability values were obtained from seed yield per hectare, days to maturity, seed yield, yield per plot, hundred seed weight, number of pod length, plant height, number of primary branch per plant, days to emergence and days to flowering whereas low heritability was obtained from number of secondary branch per plant, number seed per pod and of pod per plant. Positive and highly significant correlation were reported between grain yield and yield per plot, hundred seed weights and yield per plot while negative and significant correlation was obtained between pod length and yield per plot. Thus, genetic evaluation in these genotypes indicated that there were genotypic and phenotypic variation, positive and significant correlation and moderate to high heritability in the most studied traits that will be utilized in the future breeding program. Finally, this investigation should be repeated over years and locations to confirm future breeding program.

Key words: *Cicer arietinum*, correlation, genetic variation, heritability.

INTRODUCTION

Chickpea is the third most important pulse crop after faba bean and haricot beans in production and area coverage with annual production of 225,607.53 hectares and average productivity of 19.69 Qt./ha in the country (CSA, 2016/2013). On the contrary, in areas where improved chickpea technologies were adopted and used, yield

*Corresponding author. E-mail: kewetimieso@yahoo.com. Tel: +251910761170.

levels of up to five tons per hectare have been achieved (Tabikew et al., 2009). This huge productivity gap warrants wider dissemination of the improved chickpea technologies in order to substantially boost up the overall productivity and production in the country.

The principal uses of chickpea in Ethiopia include: as good source of protein (20 to 30%) as compared with cereals (8 to 10%), about 40% carbohydrates and 3 to 6% oil (Gil et al., 1996). It is also a good source of calcium, magnesium, potassium, phosphorous, iron, zinc and manganese (Ibrikci et al., 2003). Two main types of chickpea cultivars are grown globally: the desi and kabuli types. The total yield production is quite low in most chickpea growing countries and a wide gap exists between the potential (5 ton ha^{-1}) and actual (0.96 ton ha^{-1}) yields (FAOSTAT, 2013). The low yields have been attributed to low genetic diversity of cultivated chickpea for yield and yield components (Malik et al., 2014) and various biotic (Aschochyta blight, Fusarium wilt, Pod borer dry root rot etc.).

Plant breeders are continuously engaged to improve the genetic yield potential of this crop to meet the demands of ever-increasing population. Heritability explains whether the differences observed among individuals arose because of differences in genetic makeup or due to environmental factors. Genetic advance gives an idea of possible improvement of new population through selection, when compared to the original population. The information on nature and magnitude of genetic variation in quantitative characters and their inter-relationship in population comprising diverse genotypes is an important prerequisite for systematic breeding program. Therefore, several research workers (Malik et al., 1983; 1988; Saleem et al., 2002a, b; Parshuram et al., 2003; Ali et al., 2008) have emphasized the utility of the estimates of genetic components in the response prediction of quantitative characters to selection as well as the correlated response of various traits to grain yield.

To formulate proficient breeding program and for developing high-yielding varieties, it is essential to understand the genetics of the yield and related traits. It is recognized that, correlation coefficient indicates relation between any two traits. In order to tracing any possible causes of correlation between seed weight per plant and other yield related traits, correlation coefficient is calculated.

Selection criteria based on yield components would be helpful than direct selection in selecting suitable plant types as yield is quantitative trait that is affected by environmental variation. Thus, construction of selection indices will be highly helpful to discriminate desirable genotypes. The discriminant function provides an efficient method for simultaneous selection (Smith, 1936).

For this reason, to estimate expected genetic gain of the character through discriminant function methods is necessary. This method has been successfully followed by various researchers in various crops such as Deb and Khaleque (2007) in chickpea, Ferdous et al. (2010) in bread wheat, Kumar et al. (2012) in Rabi sorghum and Sarker et al. (2013) in chickpea. Hence, available information will be very helpful for an efficient selection criterion in selecting the most desirable and high yielding genotypes of chickpea.

Genotypic and phenotypic variances make available the information of variability only but the heritable portion of this variation is determined by the estimates of heritability. Genetic structure of the breeding materials determines the extent of heritability of various characters (Kahirizi et al., 2010). For that reason, awareness of these values of the resources in which breeders are paying attention is of enormous importance. High heritability estimates signify the effectiveness of these characters through selection for crop improvement, as less environmental influences are involved in it (Maniee et al., 2009). Malik et al. (2011) reported highest heritability together with high genetic advance expressed as percent average in 100 seed weight, seed volume and swelling index suggested effective selection for these characteristics.

The most common chickpea genotypes; Minjar, Natoli, Ejere, Mestawel, Fetenech and Cheffe (local variety) were selected to study their genetic variability. In spite of all the multi benefits of chickpea, its productivity has remained low in Ethiopian agriculture. Reasonable grain yield in chickpea could be achieved by using improved genotypes with appropriate agronomic practices. There was no research work that has been done so far on genetic variation of chickpea genotypes and correlation among yield and related traits in West Guji zone Gedeo zone.

The present investigation was to determine the genetic variation of some selected chickpea (*Cicer arietinum L*.) genotypes and correlation among yield in related Abaya areas; to estimate genetic advance and heritability of chickpea genotypes and to evaluate the correlation among yield and related traits for some selected chickpea (*C. arietinum L*.) genotypes.

MATERIALS AND METHODS

Description of experimental site

The experimental site was located in southern Ethiopia in the Oromia Regional State and the study was conducted at local farmers land, in Abaya sub site. Abaya is found in southern Ethiopian Rift valley 365 km away south from Addis Ababa. It is situated in Kolla (70%) and Woina Dega (30%). Abaya is bordered on the south by Gelana district, on the north by Gedeo Zone, on the east by Lake Abaya and on the west by Yirga Chafe District. The altitude of the district ranges from 1200-2060 m.a.s.l. Abaya receives annual rainfall ranging from 700 to 11000 mm and the average annual temperature ranges from 16 to 28°C. The rainy season occurs from April to October and the maximum rain is received in the months of May (Abaya Agricultural Office Profile and GPS readings taken from the site, 2017).

Treatments and experimental design

Some released chickpea genotypes were obtained from Debre Zeit Agricultural Research Center. A field experiment was conducted to study growth and yield of six chickpea genotypes (Ejere, Fetenech, Mestawel, Minjar, Natoli and local variety/Cheffe) evaluated in Chromic Luvisols, Eutric Fluvisols and Dystric Nitosols (Tadesse, 2002) of Abaya wereda at Guangoa Badiya site. The experiment was laid out in a randomized complete block design with three replications and the net plot size was 2.0 ×1.5 m = 3 m^2; 30 cm x 15 cm of inter and intra-row spacing; compromising of 5 rows with 13 seeds per rows were used and seedbed was prepared by ploughing 3 times followed by plantings.

Traits evaluated

The data for the following traits were recorded from ten randomly selected plants from each experimental plot, and the average value was considered: plant height, number of primary branches per plant, number of secondary branches per plant, number of pods per plant, number of seeds per pod, pod length, days to 50% flowering, days to 95% maturity, 100 Seed weight, and seed yield per plot.

Statistical analysis

Analysis of variances

All collected data were subjected to analysis of variance using appropriate computer software (SAS, 2004) and Duncan's Multiple Range Test (DMRT) at probability of 0.05 was used to separate the means and ranges for significant parameters.

Phenotype and genotype

The variability was estimated using range, mean, standard error, phenotypic and genotypic variance and coefficient of variation and the resulting components of variances were used to compute the phenotypic and genotypic variation and genetic advances as:

$\sigma^2_g = (\sigma^2_t - \sigma^2_e)/r$

Where, σ^2g = genotypic variance, σ^2t= mean square of treatment and σ^2e =error mean square, r = number of replication.

$\sigma^2_P = (\sigma^2_g + \sigma^2_e)$

Where, σ^2p = phenotypic variance.

According to Singh and Chaudhary (1999), the phenotypic and genotypic coefficients of variances (GCV) are expressed by the following formula:

GCV (%) = $(\sqrt{\sigma^2_g}/X)$ x100

X = Mean value of the trait and PCV (%) = $(\sqrt{\sigma^2_p}/X)$ x 100,

Where, PCV = phenotypic coefficient of variation

Heritability in broad sense was calculated for each trait by using the following formula (Allard, 1960):

H (%) = (σ^2_g/σ^2_p) x100

The expected genetic advance (GA) under selection, assuming the selection intensity of 5% was calculated as proposed (Johanson et al., 1955):

GA = K. $(\sqrt{\sigma^2_p})$. σ^2_g/σ^2_p = K * H * $\sqrt{\sigma^2_p}$ = K * H * σ (Standard deviation), K = standardized selection differential (K = 2.06 at 5% selection intensity).

GAM = (GA/ X) x100

Where, GAM = Genetic expected mean.

Correlation coefficient

Both genotypic and environmental effects, which are the inherent correlation between two variables, were estimated by the formula of (Al- Jibouri et al., 1958):

r_g= Gcovx.y/ $\sqrt{(\sigma^2_{gx}.\sigma^2_{gy})}$

Where, r_g=genotypic correlation coefficient, Gcovx.y =genotypic co-variance between variables x and y, σ^2_{gx} =genotypic variance for variable x and σ^2_{gy}= genotypic variance for variable y.

RESULTS AND DISCUSION

Phenotypic and genotypic coefficient

Success of plant breeders in selecting genotypes that produces higher yield and quality traits depends on existence and exploitation of genetic variability to the fullest extent. Estimation of phenotypic coefficient of variation showed that environment does have significant effect on the studied traits. The lower phenotypic and genotypic coefficient of variations were recorded from plant height, number of pod per plant, pod length, hundred seed weight and yield per pod (Table 1). The difference between phenotypic and genotypic coefficient of variations revealed that there was a little environmental influences on the traits except for seed yield per hectare, number of secondary and primary branches per plant. These results are in confirmation with those of Malik et al. (2011) and Lokare et al. (2007) who reported little influence of environment on seed physiochemical traits in chickpea.

Heritability in broad sense

Phenotypic and genotypic variances make information available for the variability only but the heritable portion of this variability was determined by the estimates heritability. Broad sense heritability (H), an estimate of the total contribution of the genotypic variance to the total phenotypic variance ranged from 34.06% for number of secondary branch per plant to 97.36% for yield per plot (Table 1).

All traits had high heritability estimates as followed for yield per plot (97.36%), days to maturity (95.51%), pod length (93.83%), 100 seed weight (87.63%), seed yield per hectare (86.92%), plant height (84.52%), days to emergence (77.46%), days to flowering (77.12%), and number of primary branch per plant (75.21%). Highest

Table 1. Estimates of genetic variability in chickpea genotypes.

Traits	Range	± SE	Mean	(σ^2 p)	(σ^2g)	(σ^2e)	PCV	GCV	H%	GA	GA as (%)
DE	1.1-2.01	1.15	1.56	23.47	18.18	5.29	6.21	5.47	77.46	7.72	9.89
DF	50.33-54.83	1.17	52.58	23.82	18.37	3.45	6.18	5.43	77.12	7.74	9.80
DM	50.5-85.67	0.70	68.09	43.62	41.66	1.96	3.92	3.83	95.51	12.97	7.69
PBPP	41.17-46.50	1.83	43.84	54.25	40.80	13.45	4.17	3.62	75.21	11.39	6.45
SBPP	41-44.17	2.69	42.59	43.86	14.94	28.92	7.47	4.36	34.06	4.64	5.23
PH	42.50-55.50	0.12	49.00	0.39	0.33	0.06	45.44	41.77	84.52	1.08	78.95
PPP	22.75-70.67	0.97	46.71	7.32	3.55	3.77	15.06	10.49	48.48	2.70	15.01
PL	7.5-12.50	0.13	10.00	1.14	1.07	0.07	24.60	23.83	93.83	2.06	47.47
SPP	3.07-3.30	1.35	3.19	14.03	6.69	3.74	10.98	7.59	47.68	3.67	10.77
HSW	14.00-16.17	0.34	13.49	3.72	3.26	0.46	14.29	13.38	87.63	3.47	25.75
YLD	180.24-437.42	1.91	308.83	112.04	97.39	14.65	2.62	2.44	86.92	18.92	4.68
YPP	11.08-17.96	0.12	14.52	2.27	2.21	0.06	16.93	16.70	97.36	3.02	33.89

DE days to emergence; DF, days to flower; DM days to maturity; PBPP, number of primary branch per plant; SBPP, number of secondary branch per plant; PH, plant height; PPP, number of pod per plant; PL, pod length; SPP, number of seed per plant; HSW, hundred seed weight; YLD, grain yield; YPP, yield per plot.

heritability of 97.36 for yield per plot along with high genetic advance indicated that maximum improvement by selection could be possible considering this trait whereas relatively moderate estimates of heritability were recorded from number of pod per plant (48.48%), number of seed per pod (47.68%), and number of secondary branch per plant (34.06%) indicating that all the studied traits may positively respond to phenotypic selection (Table 1).

High heritability estimates signify the effectiveness of these traits through selection for crop improvement, as less environmental effects were involved in the traits (Maniee et al., 2009). In addition, the current observations were in agreement with the findings of Malik et al. (2011) genetic analysis of physiochemical traits in chickpea; the highest heritability was obtained from 100 seed weight (99%) followed by seed volume (95%), swelling index (91%) indicating additive genetic variation was the major component of genetic variation in the inheritance of these traits and the effectiveness of selection in the early generation.

Estimates of expected genetic advance

The genetic advances as percent of the mean (GAM) at 5% selection intensity is presented in Table 1. It ranged from 4.68% for seed yield to 78.95 % for plant height (Table 1). This showed presence of high genetic variability in the case of plant height; but most traits showed low to moderate genetic advance for these parameter, which were also reflected by their respective low genotypic and phenotypic variations. This in turn showed the importance of genetic variability for the improvement of the traits through selection. The current observations are in confirmation with the findings of Malik et al. (2011) who reported similar results in their study on genetic analysis of physiochemical traits in chickpea genotypes.

Estimates of expected genetic advance

The genetic advances as percent of the mean (GAM) at 5% selection intensity is presented in Table 1. It ranged from 4.68% for grain yield to 78.95% for plant height to 4.68% for grain yield (Table 1). This showed the presence of high genetic variability in the case of plant height; but most traits showed low to moderate genetic advance for these parameter that are also reflected by their respective low genotypic and phenotypic variations. This in turn showed the importance of genetic variability for the improvement of the traits through selection. The current observations are in confirmation with the findings of Qurban et al. (2011) who reported similar results in their study on maize genotypes.

Correlation coefficient of six chickpea genotypes

Days to emergence showed positive and significant to highly correlations with number of pod per plant, number of seed per pod, hundred seed weight, and grain yield. Positive and significant correlations were observed between number of secondary branch per plant and yield per pod and hundred seed weights. Positive and highly significant correlation were reported between grain yield and yield per plot, hundred seed weights and yield per plot, while negative and significant correlation was obtained between pod length and yield per plot (Table 2). Similarly, Qurban et al. (2011) showed positive and significant correlation of anthesis-silking interval with ear

Table 2. Genotypic coefficient of variation in chickpea genotypes.

Traits	DE	DF	DM	PBPP	SBPP	PH	PPP	PL	SPP	HSW	YLD	YPP
DE	1.00	0.11	-0.01	-0.14	0.19	0.15	0.24*	-0.01	0.21*	0.30**	0.39**	0.16
DF		1.00	-0.07	-0.56	0.21	-0.11	0.10	-0.06	-0.06	0.05	0.10	-0.02
DM			1.00	0.07	0.07	-0.01	0.01	0.00	-0.12	0.07	-0.06	0.03
PBPP				1.00	-0.07	0.15	-0.05	-0.05	-0.13	0.05	0.01	-0.02
SBPP					1.00	0.04	0.06	0.04	0.10	0.13	0.15	0.25*
PH						1.00	0.05	-0.16	0.20*	0.26*	0.03	0.15
PPP							1.00	-0.10	-0.08	0.35**	0.47**	0.40**
PL								1.00	-0.01	0.02	-0.09	-0.25*
SPP									1.00	0.03	0.01	0.05
HSW										1.00	0.48**	0.32**
YLD											1.00	0.33**
YPP												1.00

height. Sharanappa et al. (2014) and Saurabh et al. (2017) also reported similar results.

Conclusion

The ranges of mean values for most of the traits were large showing the existence of variation among the tested genotypes. Phenotypic coefficients of variation (PCV) were found to be higher than genotypic coefficients of variation (PCV) for all the traits. The two values differed slightly indicating less influence of the environmental factors.

Moderate heritability values were obtained for days to maturity, days to flowering, number of primary branch per plant, yield per plant, number of secondary branch per plant, plant height, pod length, number of pod per plant, days to emergence, grain yield per hectare, whereas number of seed per pod had low heritability and hundred seed weights showed very low heritability. Positive and highly significant correlation were reported between grain yield and yield per plot, hundred seed weights and yield per plot, while negative and significant correlation was obtained between pod length and yield per plot.

Genetic evaluation in these genotypes indicated that there were genotypic and phenotypic variation, positive and significant correlation as well as high heritability in the most studied traits that will be utilized in the future breeding program. The study has to be repeated over years and location by increasing number of genotypes to confirm the future breeding programs.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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REFERENCES

Ali M, Nawab N, Rasool G, Saleem M (2008). Estimates of variability and correlations for quantitative traits in *cicer arietinum.* Journal of Agriculture and Social Sciences 4:177-179.

Allard RW (1960). Principles of Plant Breeding. John Willey and Sons, Inc. New York 485 p.

Al-jibouri HA Miller PA, Robinson HE (1958). Genotypic and environmental variances in an upland cotton crosses interspecific origin. Agronomy Journal 50: 633.

Central Statistical Authority (2013). Agricultural sample survey 2012/2013. Report on area and production of major crops (private peasant holdings, Meher season). Addis Ababa 19 p.

Central Statistical Authority (2016). Chickpea Production, Technology Adoption and Market Linkages in Ethiopia: Pan-African Grain Legume and World Cowpea Conference Livingstone-Zambia.Feb 28 – Mar 4, 2016.

Deb AC, Khaleque MA (2007). Study of discriminant function selection in chickpea (*Cicer arietinum* L.). Indian Biologist 39(1):51-60.

Ferdous MF, Samsuddin AKM, Hasan D, Bhuiyan MMR (2010). Study on relationship and selection index for yield and yield contributing characters in spring wheat. Journal of Bangladesh Agriculture University 8(2):191-194.

Food and Agricultural Organization of the United Nations (FASOSTAT). (2013). [online] Available at http://faostat.fao.org/site/339/default.aspx, 2013

Gil J, Nadal S, Luna D, Moreno MT, Haro AD (1996). Variability of some physico-chemical characters in desi and kabuli chickpea types. Journal of Science and Food Agriculture 71:179-184.

Ibrikci H, Knewtson SJB, Grusak MA (2003). Chickpea leaves as a vegetable green for humans: evaluation of mineral composition. Journal of the Science of Food and Agriculture 83:945-950.

Johanson N, Robinson H, Comstok R (1955). Estimates of genetic and environmental variability in soybean. Agronomy Journal 47:314-318.

Kahirizi D, Maniee M, Mohammad R, Cheghamirza K (2010). Estimation of genetic parameters related to morpho-agronomic traits of durum wheat (*Triticum turgidum var. durum*). Biharean Biologist 4:93-97

Lokare YA, Patil JV, Chavan UD (2007). Genetic analysis of yield and quality traits in kabuli chickpea. J. Food Legumes, 20(2), 147-149.

Malik B, Hussain S, Haqqani A, Chaudhry A (1983). Genetic variability in mung bean (*Vigna radiata*). Pakistan Journal of Agriculture Research48:729-735.

Malik B, Khan I, Malik M (1988). Genetic variability and correlations among metric traits in chickpea. Pakistan Journal of Agriculture

Research 9:352-354.
Malik SR, Muhammad S, Umer IM, Ashraf Z, Ahmad Bakhsh, Iqba SM (2011). Genetic Analysis of Physiochemical Traits in Chickpea (*Cicer arietinum*) Seeds. International Journal of Agriculture and Biology 13(6):1033-1036.
Malik SR, Shabbir G, Zubur M, Iqball SM, Ali A (2014). Genetic Diversity Analysis of Morpho-Genetic Traits in Desi Chickpea (*Cicer arietinum* L.). International Journal of Agriculture and Biology 16:956-960.
Maniee M, Kahrizi D, Mohammadi R (2009). Genetic variability of some morpho-physiological traits in durum wheat (*Triticum durum Desf*.). Journal of Applied Science 9:1383-1387.
Parshuram S, Mishra P, Pattnaik R, Sial P (2003). Studies on genetic variability, heritability and genetic advance in chickpea (*Cicer Arietinum* L). Environmental Ecology 21:210-213.
Qurban A, Muhammad A, Muhammed HNT, Mehboob E, Jehanzeb F, Muhammad W, Muhammad S (2011). Genetic variability for grain yield and quality traits in chicpea (*Cicer arietinum*.L). International Journal for Agro Veterinary and Medical Sciences 5(2):201-208.
Saleem M, Shahzad K, Javid M, Rauf S (2002a). Heritability estimates for grain yield and quality characters in chickpea (*Cicer Arietinum*). International Journal of Agriulture and Biology 4:275-276.
Saleem M, Tahir M, Kabir R, Javid M, Shahzad K (2002b). Interrelationships and path analysis of yield attributes in chickpea (*Cicer Arietinum* L.). International Journal of Agriulture and Biology 4:404-406.
Sarker N, Samad MA, Azad AK, Deb AC (2013). Selection for better attributes through variability and discriminant function analysis in chickpea (*Cicer arietinum* L.). Journal of Subtropical Agricultural Research and Development 11(1):1050-1055.
Saurabh S, Roopa LG, GM Lal (2017). Genetic variability and character correlation for seed yield in chickpea (*Cicer arietinum* L.). Journal of Pharmacognosy and Phytochemistry. 6(4):748-750.
Sharanappa SD, Kumar J, Meena HP, Bharadwaj C, Jagadeesh HM, Raghvendra KP, Singode A (2014). Studies on heritability and genetic advance in chickpea (*Cicer arietinum* L.). Journal of Food Legumes 27(1):71-73.
Singh RK, Chaudhary BD (1999). Biometrical Genetics Analysis, Kalyani Publishers, New Delhi 318P.
Smith HF (1936). A discriminant function for plant selection. Annals of Eugenics 7: 240-250.
Statistical Analysis System (2004). SAS user guide, statistics SAS Inc. Cary. Northern Carolina, USA.
Tabikew D, Asnake F, Kibebew A, Lijalem K (2009). Improved Chickpea (*Cicer arietinum* L.)Technologies and seed production in Ethiopia. Ethiopia Agricultural Research Center Institute (EIARC), Research bulletin, Debre Zeit, Ethiopia.

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