Genetic Diversity Studies on Yield and Its Related Traits in Korarima (Aframomum Corrorima (Braun) Jansen)) Germplasms

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Genetic diversity studies was undertaken on 25 korarima germplasm collected from different agro ecological areas of Ethiopia to help in identifying elite germplasm accession with the greatest novelty at Jimma agricultural research center during the year of 2011 from August to December. The experiment was superimposed on those which were planted in a 5x5 simple lattice design with two replications and five accessions per incomplete block. Nine plants per plot were planted with a spacing of 1.8m both between rows and plants. The germplasms were grown under Sesbania shade trees. Cluster analysis revealed that the 25 korarima germplasm were grouped in to four clusters. Distance among these clusters is significantly different for all the cluster combination. This indicates that there is an opportunity to bring about improvement through hybridization of germplasm from different clusters and subsequent selection from the segregating generations. Principal component analysis indicated that six principal components explained about 80.51% of the total variation. Differentiation of germplasms into different cluster was because of cumulative effect of number of characters. The present study generally implied the presence of significant genetic variability among the tested genotypes. Thus, there is an excellent opportunity to bring about improvement through direct selection and hybridization which involves crossing of genotypes from different clusters.

Keywords: korarima (Aframomum corrorima), genetic diversity, genetic divergence, yield per plant.

1. Introduction

Korarima (Aframomum corrorima) or Ethiopian cardamom is herbaceous, perennial and aromatic spice and medicinal crop of the species in the monocotyledonous ginger family, Zingiberaceae native to Ethiopia. It is a shade loving plant that grows wild in moist and open woodlands, in the same climate areas as wild coffee, but may also be planted and cultivated. The plant consists of an underground rhizome, a pseudostem, and several broad leaves and resembles Elettaria species morphologivally Jansen(1981).

The seeds of korarima contain different types of essential oils having typical odour (Jansen 1981; Abegaz *et al.*,1994; Eyob *et al.*,2007) and are traditionally used as tonic, carminative and purgative drug. From a formal survey, korarima seeds, pods, leaves, rhizomes and flowers are used in southern Ethiopia as traditional medicine for human and animal ailments caused by unknown agents; and particularly used to treat any part of the animal body upon swelling (Eyob *et al.*,2008).

Korarima seed has a mild, sweet flavour and is less peppery or pungent than seed of *Aframomum melegueta* K.Schum. (grain of paradise). The seeds contain essential oil which has a typical odour and is sometimes called 'nutmeg-cardamom'. After distillation of dried comminuted fruits, 3–3.5% of a pale yellow volatile oil with a flat cineolic odour can be obtained, in which the following compounds have been found (all monoterpenes, approximate amount of the major ones): 1,8-cineol 32–35%, limonene 7–14%, β-pinene 4–7%, sabinene 7–9%, terpinen-4-ol 3–5%, geraniol 5%, P-cymene 4%, α -pinene, α -terpineol and γ -terpinene 3% each. Sesquiterpenes were identified in another analysis; the total was dominated by about 75% monoterpenes including 1,8-cineol (38%) and terpinyl acetate (11%), and 17% sesquiterpenes including nerolidol (11–14%), β-caryophyllene (2%) and caryophyllene oxide 1% (Eyob *et al.*, 2007).

Assessment of genetic variability in crop has a strong impact on plant breeding and conservation of genetic resources (Van Hintum, 1995). It is particularly useful in the characterization of individuals, accessions, and cultivars in order to determine the level of genetic diversity available in germplasm collections and for selecting parents. The prior knowledge of the nature, extent and distribution of genetic variation is crucial for successful conservation (*in-situ* and *ex-situ*) and sustainable utilization of germplasm.

Moreover, the number of populations necessary to conserve genetic diversity within a species and choice of sites for *in-situ* conservation depend on the measure of diversity and its pattern of partition within and among populations (Kassahun, 2006).

Currently, under Ethiopia korarima improvement project, large numbers of korarima accessions are collected from different major growing regions of Ethiopia by Jimma Agricultural Research Center (JARC). As

far as the genetic diversity study in these accessions of korarima is concerned nothing has been done. Hence, the present study was undertaken to estimate genetic diversity among accessions collected from different agro ecological areas of Ethiopia.

2. Materials and methods

Description of the Study Areas

The study was carried out at Jimma Agricultural Research Centre located at 363 km south west of Addis Ababa. The site is located at 7046' N and 360 E with an altitude of 1753 meters above sea level. It is situated in the tepid to cool humid-mid highlands of southwestern Ethiopia. The area is among the most conducive production areas for *aframomum corarima* in Ethiopia and the accessions were expected to fully express their genetic potential for the trait under consideration. The soil type of the experimental area is Eutric Nitosol (reddish brown) with a pH of around 5.2. The area receives mean annual rainfall of 1536 mm with a maximum and minimum temperature of 25.9 o C and 11.2 o C, respectively (IAR, 1997).

Experimental Materials

The materials used for the study were 25 already established Ethiopian korarima germplasm accessions that are five years old and local checks. The korarima germplasm accessions were collected from the potential and representing areas. The list of geneotypes used in the study is given in Table 1. Table 1 germplasms used in the experiment

No	Accession	Region	Zone	Woreda	Altitude
					(m. a.s.l)
1	Jimma local	Oromia	Jimma	Jimma	1580
2	028/84	Oromia	Wollega	Arjo	1800
3	025/03	Oromia	Illubabor	Metu	1605
4	114/03	Oromia	Illubabor	Sombo	2229
5	059/03	Oromia	Wollega	Nekemte	2088
6	029/84	Oromia	Wollega	Gimbi	1930
7	016/84	Oromia	Illubabor	Sombo	2229
8	001/03	SNNPR	Sheka	Masha	1297
9	015/03	Oromia	Ilubabor	Sombo	2229
10	053/03	SNNPR	South Omo	Kemba	1850
11	045/03	SNNPR	Gamo gofa	Damot	2121
12	701/87	SNNPR	Kefa	Decha	2500
13	046/03	Oromia	Illubabor	algea	1500
14	105/03	Oromia	Illubabor	Yayu	1387
15	038/01	SNNPR	Sidama	Arero	2829
16	093/00	Amhara	Gojam	Debremarkos	2446
17	018/00	SNNPR	Kefa	Yeki	1097
18	010/00	SNNPR	Kefa	Chena	1972
19	009/00	Amhara	Gojam	Metekel	1525
20	068/87	Amhara	Gojam	Agew midir	500-3700
21	021/00	SNNPR	Bench maji	Bebeka	950-1285
22	686/87	Amhara	Gojm	Metekel	1525
23	001/84	Oromia	Bale	Genale	1000
24	011/00	SNNPR	Sidama	Sidama	2759
25	014/00	Amhara	Gojam	Metekel	1525

Source: Jimma Agricultural Research Center

Experimental Design, Management and Season

The study was conducted in 2011 from August to December. The experiment was superimposed on those which were planted in a 5x5 simple lattice design with two replications and five accessions per incomplete block. Nine plants per plot were planted with a spacing of 1.8m both between rows and plants. The genotypes were grown under Sesbania shade trees.

Data collected

Five plants were randomly selected from each plot to take average measurements for parameters like, plant height (cm), number of tiller per plant, number of bearing tiller per plant, intermodal length (cm), number of leaves per stem, leaf area (cm²), number of capsule per plant and yield per plant. For weight of single capsule(g), length of single capsule(cm) and diameter of single capsule(cm) both at fresh and dry base were measured by taking twenty five capsule from each five randomly selected plant of plots. Essential oil and oleoresin extraction

was carried out by the procedure of ASTA(1997) and dry matter, total ash, crud fiber and crud fat content on percent base were determined by the procedure of A.O.A.C,(1990).

Data analysis

Data of quantitative characters were subjected to analysis of variance (ANOVA) using SAS version 9.2 (SAS, 2008) to examine the presence of statistically significant differences among genotypes for these characters. Clustering was performed using the proc cluster procedure of SAS version 9.2 (SAS institute, 2008) by employing the method of average linkage clustering strategy of the observation. The numbers of clusters were determined by following the approach suggested by Copper and Miligan (1988) by looking in to three stastics namely Pseudo F, Pseudo t^2 and cubic clustering criteria. Principal component analysis was performed using correlation matrix by employing SAS procedure (SAS, 2008). The objective of this analysis was to reduce the observed variables in smaller number of principal component that were accounted for most of the variance in the observed variables. Finally, it defines the pattern of variation between the accessions by summarizing data in to reduced number of traits (Corossa *et al.*, 1995)

Analysis of variance (ANOVA)

Mean squares of 21 characters from analysis of variance (ANOVA) presented in table 2. Significant difference among germplasm accessions (p<0.05) were observed for all traits expect for seed weight, internodal length and percent dry matter content of korarima seed. Significant difference indicates the presence of variability. Different authors reported significance difference on different characters of cardamom and other crop genotypes. From those Korikanthimath *et al.*(2000) reported significance difference among genotypes for number of capsule per plant, weight of fresh and dry capsule and oleoresin content. Ankegowda and Krishnamurthy (2008) also reported number of tiller, number of leaves and plant height show significant difference on six cardamom germplasm accessions under moisture stress condition which is in line with this finding.

SV	REP	Treatments	B/REP	ERROR		R ²⁰ %	CV%
				Intra block	RCBD		
DF	1	24	8	16	24		
PH (cm)	383.09	669.76**	104.63	276.28	219.06	79.29	8.68
TT	0.5202	5.93**	2.87	2.293	2.48	82.42	19.89
BT	0.39	1.24**	0.56	0.22	0.33	88.42	20.93
INL (cm)	0.004	0.52 ^{NS}	0.23	0.45	0.38	60.28	13.35
NLPS	1.095	19.52**	9.49	7.27	8.01	81.78	9.32
LA (cm ²)	61.16	2826.66**	215.76	919.69	685.05	84.71	16.95
NCPP	0.08	4.06**	0.8	1.26	1.1	83.37	12.43
YPP(g)	244.3	5885.59**	1393.1	2014.88	1807.62	82.5	14.47
SW (g)	8.82	0.035 ^{NS}	0.014	0.023	0.02	69.29	7.03
WFC (g)	0.014	28.46*	16.12	12.54	13.73	75.81	14.49
LFC (cm)	0.03	6.065*	3.64	2.85	3.11	78.16	20.3
DFC (cm)	3.28	4.23**	2.08	0.95	1.33	83.48	9.81
WDC (g)	0.23	9.36*	6.45	3.32	4.36	77.16	15.78
LDC (cm)	1.4	1.24*	0.32	0.59	0.5	81.37	16.39
DDC (cm)	3.28	4.23**	2.08	0.95	1.33	83.48	15.48
DRM(%)	0.61	1.41 ^{NS}	0.203	1.41	1.62	62.04	1.37
CRFI (%)	0.0006	0.0048**	0.0014	0.0011	0.0011	86.41	15.79
VOC (v/w)	0.23	0.53**	0.23	0.19	0.21	87.53	17.93
OC (w/w)	1.48	1.147**	0.403	0.365	0.378	83.31	12.03
ASH (%)	0.021	0.48**	0.16	0.146	0.149	89.82	15.36
CRFAT(%)	0.027	0.051**	0.037	0.021	0.026	81.32	5.69

** and * indicates significant difference at 1 and 5% respectively, NS not significant

DF: degree of freedom, PH: plant height, TT:total tiller, BT: bearing tiller, IL: internodal length, NLPS: number of leaf per stem, LA: leaf area, NCPP: number of capsule per plant, YPP: yield per plant, SW: 100seed weight, WFC: weight of fresh capsule, LFC: length of fresh capsule, DFC: diameter of fresh capsule, WDC: weight of dry capsule, LDC: length of dry capsule, DDC: diameter of dry capsule, DRM%: dry matter percentage, CRFI%: crud fiber percentage, VOC: volatile oil content, OC: oleoresin content, %ASH: percent ash content, CRFAT%: crud fat percentage

Cluster analysis

Cluster analysis grouped 25 accessions in to four distinct groups (Table 3) in which the first cluster (CL1) consisted of 9 accession which is (36%), the second cluster (CL2) 6 accessions (24%), the third cluster (CL3) 8 accessions (32%) and fourth cluster (CL4) containend 2 accession (8%).

Table 3. Distribution of	of germplasm acces	ssion in to fo	our clusters based	on D^2 analysis	for 25 korarima
accessions studied at J	ARC 2011/12				

Clusters	Number germplasm	serial number	Region
	accession		
Cluster I	9	015/03	Oromia
		053/03	SNNPRS
		068/87	Amhara
		701/87	SNNPRS
		010/00	SNNPRS
		028/84	Oromia
		105/03	Oromia
		001/03	SNNPRS
		046/03	Oromia
Cluster II	6	029/84	Oromia
		011/00	SNNPRS
		045/03	SNNPRS
		001/84	Oromia
		114/03	Oromia
		059/03	Oromia
Cluster III	8	Jimma local	Oromia
		093/03	Amhara
		025/03	Oromia
		009/00	Amhara
		016/84	Oromia
		038/01	SNNPRS
		021/84	SNNPRS
		686/87	Amhara
Cluster IV	2	018/00	SNNPRS
		014/00	Amhara

Cluster mean analysis

Collections from SNNP regional states were almost distributed in all clusters than Amhara and Oromia regional states indicating the existence of more genetic diversity in this region than Amhara and Oromia regional states and accession from the same origin might have different genetic background.

Cluster I characterized by having the tallest plant, highest weight of fresh capsule, length of fresh capsule, diameter of dry capsule, oleoresin and ash contents. It also showed lowest number of total tiller and bearing tiller per plant, length of dry capsule and crud fiber content.

Cluster II was characterized by the highest volatile oil and crud fat content but with lowest number of leaves per stem, yield per plant diameter of fresh and dry capsule.

Cluster III exhibited by the highest total tiller, number of leaf per stem, weight of dry capsule, length of dry capsule and crud fat content. It also exhibited the lowest leaf area and number of capsule per plant.

Cluster IV was characterized by the highest yield, number of bearing tiller, leaf area and number of capsule per plant. It was characterized by shortest plant, the lowest weight of fresh capsule, length of fresh capsule, weight of dry capsule, volatile oil, oleoresin, ash and crud fat content.

Table 4. Mean value of 18 quantitative characters of the four clusters for 25 korarima	germplasm accession
studied at JARC 2011/12	•

Traits	cluster	rs		
	I II	III	IV	
PH	185.6**	166.8	170.9	144.6*
TT	6.4*	7.4	7.7**	6.9
BT	2.2*	2.4	2.8	3**
NLPS	26.2	24*	30**	29
LA	194.4	132.5	131.6*	226.5**
NCPP	8.3	8.1	7.8*	9.1**
YPP	255.7	219.1*	330.3	345.3**
WFC	23.8**	22.3	23.5	20.4*
LFC	7.9**	7.8	7.4	6.6*
DFC	11.5**	10.1*	11.1	10.2
WDC	11.9	11.6	12.1**	9.5*
LDC	3.9*	4.1	4.7**	4.6
DDC	7.5**	6.1*	7.1	6.2
VOC	2.3	2.4**	2.2	1.5*
OC	5.1**	4.8	4.3	3.8*
ASH	2.4**	2.2	2.2	1.7*
CRFI	0.18*	0.24**	0.19	0.2
CRFAT	2.4	2.4	2.5**	2.3*

** and * : highest and lowest cluster mean values respectively

PH: plant height, TT: total tiller, BT: bearing tiller, NLPS: number of leaf per stem, LA: leaf area, NCPP: number of capsule per plant, YPP : yield per plant, WFC: weight of fresh capsule, LFC: length of fresh capsule, DFC: diameter of fresh capsule, WDC: weight of dry capsule, LDC: length of dry capsule, DDC: diameter of dry capsule, CRFI: crud fiber, VOC: volatile oil content, OC: oleoresin content, ASH: ash content, CRFAT: crud fat .

Genetic divergence among accessions

The squared distance was calculated and indicated in Table 5. Test of significance show significance difference between all cluster distances. The minimum squared distance was between cluster III and IV (67.75) followed by cluster I and II (82.12). Maximum squared distance was between cluster II and IV(408.27) followed by cluster II and III(240.57) and cluster I and IV (218.75). Generally this study revealed that germplasm accessions included in this study are moderately divergent. Radhakrishnan et al. (2006) using 90 caradmom genotypes reported diversity for growth and yield attributes among the accessions and they grouped into 8 clusters. According to them inter-cluster distance values also showed wide genetic divergence among accessions

According to Ghaderi et al.(1984) increasing parental distance implies a great number of contrasting alleles at the desired loci, and then to the extent that these loci recombine in the F_2 and F_3 generation following a cross of distantly related parents, the greater will be the opportunities for the effective selection for yield factors Table 5. Generalized squared distance among four clusters in 25 korarima germplasm accession studied at JARC 2011/12

Clusters		II	III	IV	
	Ι	82.12**	111.42**	218.75**	
	II		240.57**	408.27**	
]	II			67.75**	

 x^2 significant X² = 28.87 and X² = 34.81 at 5 and 1% probability level

Principal Component Analysis

Principal component analysis is presented in Table 6 and it revealed that six principal components PCI to PCVII with eigenvalues, 4.69, 3.46, 1.84, 1.64, 1.52 and 1.32 respectively, have accounted for 80.51% of total variation. The first principal components PCI and PCII with values 26.1%, and 19.27% respectively contributed more to the total variation

According to Chahal and Gosal (2002), characters with the largest absolute values closer to unity with in the first principal component influence the clustering more than those with lower absolute values closer to zero.

Characters having relatively higher values in the first principal components (PCI) include plant height, weight of fresh capsule, length of fresh capsule, diameter of fresh capsule, weight of dry capsule and diameter of dry capsule. Total tiller, bearing tiller, number of leaf per stem, yield per plant, length of dry capsule and oleoresin content in the second principal components(PCII). Bearing tiller, number of leaf per stem, volatile oil content and crud fiber content in the third principal components. Leaf area, length of fresh capsule, volatile oil content and crud fat content in the fourth principal components (PCIV).

Leaf area, weight of dry capsule, crud fiber and crud fat content in the fifth principal components (PC5) and Leaf area, number of capsule per plant and length of dry capsule in the six principal components.

Similarly Patil *et al.*(2000) reported five principal components with eigenvalues greater than 1 explaining 94.36% of the total variation by using 13 morphological traits and dry capsule yield per clump in large cardamom. Gupta *et al.*(2006) also reported three principal components explaining 73% of total variation by using seven characters on large cardamom.

Table 6. Eigenvectors and eigen values of the first six principal components (PCs) for 18 characters of 25 korarima accessions studied at JARC 2011/12

Traits			Eigenvectors			
	PC1	PC2	PC3	PC4	PC5	PC6
PH	0.359	0.199	-0.066	-0.124	-0.107	-0.18
TT	0.223	-0.314	-0.182	0.059	-0.095	-0.043
BT	0.2	-0.343	-0.334	0.1	-0.171	0.158
NLPS	0.07	-0.311	0.389	-0.01	0.053	-0.044
LA	0.066	0.079	0.095	-0.554	-0.301	0.335
NCPP	-0.025	-0.001	-0.123	-0.123	0.274	0.722
YPP	0.076	-0.418	0.298	0.093	-0.069	0.121
WFC	0.364	0.161	0.095	0.114	0.24	0.126
LFC	0.334	0.097	0.021	-0.309	0.125	-0.254
DFC	0.37	0.151	0.222	0.124	-0.191	0.11
WDC	0.364	0.17	0.011	0.106	0.318	0.106
LDC	0.22	-0.343	0.003	-0.023	0.051	-0.325
DDC	0.37	0.151	0.222	0.124	-0.191	0.11
VOC	0.073	0.059	-0.309	0.573	-0.279	0.125
OC	-0.076	0.355	-0.188	-0.117	-0.139	-0.216
ASH	-0.068	0.296	0.008	0.224	-0.169	0.004
CRFI	0.141	-0.047	-0.441	-0.051	0.506	-0.068
CRFAT	-0.173	0.143	0.392	0.306	0.381	-0.042
Eigenvalue	4.69	3.46	1.84	1.64	1.52	1.32
% variance	26.1	19.27	10.23	9.12	8.45	7.34
cumilative	26.1	45.37	55.6	64.72	73.17	80.51

PH: plant height, TT: total tiller, BT: bearing tiller, NLPS: number of leaf per stem, LA: leaf area, NCPP: number of capsule per plant, YPP : yield per plant,WFC: weight of fresh capsule, LFC: length of fresh capsule, DFC: diameter of fresh capsule, WDC: weight of dry capsule, LDC: length of dry capsule, DDC: diameter of dry capsule, CRFI: crud fiber , VOC: volatile oil content, OC: oleoresin content, ASH: ash content, CRFAT: crud fat .

Conclusion

The present study generally implied the presence of significant genetic diversity among the tested germpasm accessions. Thus, there is an excellent opportunity to bring about improvement through direct selection and hybridization which involves crossing of genotypes from different clusters.

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