

## Genetic Variability and Distance of East Africa Cooking Banana (*Musa* sp.) Clones for Morpho-physicochemical Traits

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**Abstract:** Understanding the genetic variability and diversity of crops is the basis for breeding and improving the crops. Therefore, a field experiment was conducted at Dire Dawa, eastern Ethiopia to evaluate 11 cooking banana (*Musa* sp.) clones of East Africa origin. The clones were planted in randomized complete block design with five replications. Genetic variability and distance of the clones were studied considering 19 morpho-physicochemical traits. The differences among the clones were significant for all traits except mean hand weight. Both phenotypic and genotypic coefficients of variation were moderate to high ranging from 6.67 to 57.22 and 1.21 to 47.26, respectively, with low differences in the magnitude of all traits except number of leaves at harvest, leaf breadth, and total soluble solid. A positive and more significant genotypic correlation than phenotypic correlation was observed between bunch weights with its components (mean weight of hand, number of fruits per hand and fruit weight). High heritability and expected genetic advance were detected for most of the traits, which ranged from 3.30 to 91.24 and 0.39 to 68.71%, respectively, indicating the expression of these traits, were more dependent on genetic rather than non-genetic factors. Clustering of clones resulted in two groups with one group comprising a single clone (Wendo Genet 3) while the remaining 10 forming one big group. In conclusion, the results of the study have revealed the presence of considerable genetic distances among the east African cooking banana clones. The results demonstrated that higher bunch weights and fruit yields could be achieved through indirect selection of yield components. The existence wider genetic variability among the clones can be utilized for improving the crop to enhance food production and income of farming communities.

**Keywords:** Clustering; Genotypic and Phenotypic Coefficients of Variation; Heritability.

### 1. Introduction

Bananas and plantains are the fourth most important crops in developing countries (Heslop-Harrison and Schwarzscher, 2007). Plantain and cooking bananas belong to the sub-groups AAB and ABB are important components of food security in the tropics, providing income to the farming community through local trade (Crouch *et al.*, 1998).

The modern day edible bananas are the mix of wild and cultivated species and hybrids associated with *Musa acuminata* and *Musa balbisiana*. *Musa acuminata* is the most widespread of the species (Daniell *et al.*, 2001) and the center of diversity is thought to be either Malaysia (Simmonds, 1962) or Indonesia (Horry *et al.*, 1997).

East and West Africa represent two main secondary centers of *Musa* diversity as a result of a long history of cultivation in these regions. There are approximately 60 cultivars of African highland bananas unique to East Africa, but it is not known whether these were derived from traded plants or from indigenous edible diploids (Daniell *et al.*, 2001). These highland bananas have the AAA genotype (Karamura, 1998). In this region, banana is characterized by the abundant presence of cultivars of AAA type for cooking and brewing that is not found in Asia. People in this area depend heavily on banana for the staple food. On the other hand, in

the area stretching from Central to West Africa, the plantain subgroup of ABB genome type is highly developed and plantain cultivars used for cooking play an important role as the staple food (JAICAF, 2010).

In Ethiopia, fruit crops in general and bananas in particular play an important role in the national food security. However, banana research has no long history in the country, and was started as recently as 1972 (EARO, 1999).

Banana research was focused on dessert banana to identify high yielding hybrids and cultivars for different growing regions. In Ethiopia, dessert type is a popular fruit crop. All types (dessert, cooking and beer) are available in banana growing regions of Ethiopia but all are consumed fresh (EARO, 1999). In breeding programs, for selection of genotypes with desirable traits, knowledge of nature and magnitude of variation existing in available plant breeding materials and interrelationships between quantitatively inherited plant traits is of great importance (Khan *et al.*, 2010 and Kotal *et al.*, 2010). Though the presence of variation is critical for selection in all crops, it is most important in sterile bananas which develop their fruit through parthenocarpy (Pollefeys and Arnaud, 2004). Because, the nature of these crops obstructs improvement through hybridization, exploitation of the existing

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variability is the easiest and best option to do so. Therefore, this study was conducted with the objectives of estimating, (1) phenotypic and genotypic variability, heritability of traits and genetic advance, (2) phenotypic and genotypic correlations of morpho-physicochemical traits and, (3) estimating the genetic distance of the clones.

## 2. Materials and Methods

### 2.1. Description of the Study Area

The field experiment was conducted at Dire Dawa Agricultural Research Station of Haramaya University during the 2000 to 2003 cropping seasons. Dire Dawa is located between 09° 28.1 N latitude and 41° 38.1 E longitude. The altitude of the station is 1116 meters above sea level and the mean annual temperatures range from 19. °C to 31. 5. °C. The temperature is generally high with monthly mean maximum of 28.1 °C (December) and 34.6 °C (June). Dire Dawa enjoys a bi-modal type of rainfall with April as a peak for the scanty rainfall and July for the heavy rains. The mean annual rainfall in the study area is 550 mm (Levoyageur Weather, 2012).

### 2.2. Experimental Material

Eleven cooking banana clones, namely; Wendo Genet 4, and Wendo Genet 3 from Ethiopia, Saba (AAB) Saba (AAB) (originating from the Philippines) and Kibungo I (most commonly in Rwanda), Matoke (Uganda), Cardaba, Chibule Angombe, Nijuru, Ikj manga, Cachacko, and Kitawira introduced from Kenya were used for this field experiment.

### 2.3. Treatments and Experimental Design

The treatments consisted of eleven banana clones. Suckers of the banana clones were established in randomized complete block design (RCBD) with five replications. Five plants were planted for each clone in each replication with the spacing of 2.5 meters between plants and 2.5 meters between rows. Irrigation, fertilization, weeding and other sucker management practices were applied uniformly.

### 2.4. Data Collection and Analysis

#### 2.4.1. Morphological Traits

The pseudostem height (PSH m) was measured using a tape meter from the base to the top of the plant; the girth of pseudostem (GS cm) was measured with tape meter around the circumference; the number of leaf per plant at harvest (NLH) was counted in each plant at the time of harvest; the time interval between successive leaf emergence (TBSLE) recorded in days taking into consideration the emergence day of the preceding leaf; the number of suckers up to the time of shooting (NSUTS) was recorded by counting the number of suckers produced by each plant up to the shooting time; leaf length (LL cm) was measured with the tape meter from leaf blade base to the tip of the leaf at the time of flowering for each plant; leaf breadth

(LB cm) was measured with the tape meter at the point of maximum leaf breadth at the time of flowering.

#### 2.4.2. Fruit yield and Physicochemical Traits

Bunch weight was taken using a field balance (MBW kg); the number of hands per bunch (MNH/B) was recorded after counting the hands in each bunch; the hands were separated from the bunches and weighted (MWH kg); the fingers in each hand were counted and recorded as the number of fingers per hand (MNF/H). All fingers were separated from a hand, weighted and divided to the number of fingers counted in each hand to obtain finger weight (MWF g), and fruit yield per hectare (FY/ha ton) was obtained by multiplying the number of fruits produced in a hectare by the mean weight of fingers, which was converted to ton/ha. Finger length (MFL cm) and finger diameter (MFD cm) were measured using a ruler, taking randomly selected 10 fingers from each bunch obtained from each plant in each replication. Peel weight (Mpel g) and pulp weight (Mpulp g) of fingers were measured after the peel and pulp were separated using a sensitive balance. A 30 g of pulp was cut from a pulp of banana and diluted with 90 g distilled water in a blender for one minute and filtered. A hand held refractometer was used to measure the total soluble solid (TSS) in %Brix where the reading was multiplied by three as a dilution factor. Titrable acid (TA) was measured from the diluted pulp titrated with 0.1 N NaOH to the end point at pH= 8.1.

#### 2.4.3. Data Analysis

Analysis of variance, phenotypic and genotypic variance and coefficient of variation were computed with SAS statistical software (9.0); heritability and genetic advance were computed using the excel microsoft program. Dendrogram was generated based on Unweighted Pair-group Method with Arithmetic means (UPGMA). Euclidean distances depicting genetic relationships among 11 cooking banana clones based on 19 morpho-physicochemical traits were also computed. Both UPGMA and Euclidean distances were computed using STATISTICA-7 basic statistical analysis software (U.S.A.).

The phenotypic and genotypic variance and coefficient of variation were estimated according to the methods suggested by Burton and Devane (1953) as follows:

$$\sigma^2 p = \sigma^2 g + \sigma^2 e \text{ and } \sigma^2 g = \frac{Mg - Me}{r} \dots\dots\dots(1)$$

where  $\sigma^2 p$  = phenotypic variance,  $\sigma^2 g$  = genotypic variance,  $\sigma^2 e$  = environmental variance i.e. error mean square from ANOVA,  $Mg$  = mean square of genotypes,  $Me$  = mean square of error and  $r$  = number of replication. Phenotypic coefficient of variation

computed (PCV) and genotypic coefficient of variation GCV computed as follows:

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

$$GCV = \frac{\sqrt{\sigma^2 g}}{\bar{x}} \times 100 \text{-----}(3)$$

where  $\bar{x}$  = population mean.

Heritability ( $H^2$ ) in broad sense for all traits was computed using the formula adopted by Allard (1960) and Falconer (1990) as:

$$H^2 = [\sigma^2 g / \sigma^2 p] \times 100 \text{-----}(4)$$

where,  $\sigma^2 g$  = genotypic variance,  $\sigma^2 p$  = phenotypic variance and  $\sigma^2 e$  = error variance. Genetic advance (GA) for each trait was computed using the formula adopted by Johnson *et al.* (1955) and Allard (1960) as:

$$GA = (k) (\sigma p) * (H^2) \text{-----}(5)$$

$$GA \text{ (as \% of the mean)} = \left[ \frac{GA}{\bar{x}} \right] \times 100 \text{-----}(6)$$

where,  $k$  = selection differential ( $k=2.06$  and  $1.76$  at  $5\%$  and  $10\%$ , respectively, selection intensity),  $\sigma p$  = phenotypic standard deviation,  $H^2$  = heritability in broad sense and  $\bar{x}$  = grand mean. Phenotypic and genotypic correlations between yield and yield related traits were estimated using the method described by Miller *et al.* (1958) as

$$r_{pxy} = \frac{COV_{pxy}}{\sqrt{\sigma^2 px \cdot \sigma^2 py}} \text{-----}(7)$$

Where,  $r_{pxy}$  = phenotypic correlation coefficient between character  $x$  and  $y$ ,  $COV_{pxy}$  = phenotypic covariance between character  $x$  and  $y$ ,  $\sigma^2 px$  = phenotypic variance for character  $x$  and  $\sigma^2 py$  = phenotypic variance for character  $y$ .

$$r_{gxy} = \frac{COV_{gxy}}{\sqrt{\sigma^2 gx \cdot \sigma^2 gy}} \text{-----}(8)$$

Where;  $r_{gxy}$  = genotypic correlation coefficient between character  $x$  and  $y$ ,  $COV_{gxy}$  = genotypic covariance between character  $x$  and  $y$ ,  $\sigma^2 gx$  = genotypic variance for character  $x$  and  $\sigma^2 gy$  = genotypic variance for character  $y$ .

The coefficient of correlation at phenotypic level was tested for their significance by comparing the values of correlation coefficient with tabulated  $r$ -value at  $g-2$  degree of freedom, where ' $g$ ' is no of genotypes.

However, the coefficients of correlations at genotypic level were tested for their significance using the formula described by Robertson (1959) as described below:

$$t = \frac{(r_{gxy})}{SE_{r_{gxy}}} \text{-----}(9)$$

The calculated ' $t$ ' value was compared with the tabulated ' $t$ ' value at  $g-2$  degree of freedom at  $5\%$  level of significance. Where,  $g$  = number of genotypes,  $r_{gxy}$  = genotypic correlation coefficient and  $SE_{r_{gxy}}$  = standard error of genotypic correlation coefficient between character  $x$  and  $y$  which was calculated as:

$$SE_{r_{gxy}} = \sqrt{\frac{(1-r^2)^2}{2H^2_x \cdot H^2_y}} \text{-----}(10)$$

Where,  $SE_{r_{gxy}}$  = standard error of genotypic correlation coefficient between character  $x$  and  $y$ ,  $h_x$  and  $h_y$  = heritability value of character  $x$  and  $y$ , respectively.

As Sneath and Sokal, (1973) indicated, Euclidean distance (ED) was computed from the 19 morpho-physicochemical traits of 11 cooking banana clones after standardization (subtracting the mean value and dividing it by the standard deviation) as follows:

$$ED_{jk} = \sqrt{\sum_{i=1}^n (X_{ij} - X_{ik})^2} \text{-----}(11)$$

where  $ED_{jk}$  = distance between cultivars  $j$  and  $k$ ;  $x_{ij}$  and  $x_{ik}$  = morpho-physicochemical traits values of the  $i^{th}$  character for clones  $j$  and  $k$ , respectively; and  $n$  = number of morpho-physicochemical traits used to calculate the distance.

The distance matrix from morpho-physicochemical traits was used to construct dendrograms based on the Unweighted Pair-group Method with Arithmetic means (UPGMA). The results of cluster analysis were presented in the form of dendrogram. In addition, mean ED was calculated for each clone by averaging of a particular clone to the other 10 clones. The calculated averages distance (ED) was used to estimate which clone is closest or distant to others.

### 3. Results

#### 3.1. Analysis of Variance and Mean Performance of Clones

##### 3.1.1. Analysis of Variance

The mean squares from analysis of variance for cooking banana clones with respect to 19 morpho-physicochemical traits are presented in Table 1. The results revealed the presence of significant ( $P < 0.01$ ) differences for 18 out of 19 traits studied. The overall result showed the presence of sufficient genetic variability among the tested cooking banana cultivars.

Table 1. Mean squares from analysis of variance for 19 morpho-physicochemical traits of 11 cooking banana clones.

Trait	Rep (4)	Geno (10)	Error (40)	CV (%)
Mean pseudostem height (m)	0.04*	1.448.00**	0.03	8.19
Girth of pseudostem (cm)	64.00	655.96**	21.95	6.94
Time interval between successive leaf emergence	1.07*	13.86**	0.31	7.54
Number of suckers until time of shooting	1.29	15.10**	1.12	15.09
Mean number of leaves at harvest	4.05	4.64**	1.29	8.72
Mean leaf length (cm)	418.85	2540.10**	319.43	12.04
Mean leaf breadth (cm)	18.34	91.96**	21.15	7.69
Mean bunch weight (kg)	18.48	27.26**	6.33	29.72
Mean number of hand per bunch	2.21**	3.45**	0.42	11.02
Mean hand weight (g)	134105.08	282906.52	125649.02	27.91
Mean number of finger per hand	539.56	1887.69**	406.31	26.85
Mean finger weight (g)	102.07	4565.24**	404.92	22.86
Mean peel weight (g)	179.54	746.64**	63.60	32.25
Mean pulp weight (g)	152.84	1947.03**	261.72	27.05
Mean finger length (cm)	2.34	16.75**	2.67	13.37
Mean finger diameter (cm)	0.02	0.99**	0.08	8.73
Finger yield per hectare (ton)	27.63	24.86**	14.54	32.08
Total soluble sugar	0.03	0.13**	0.11	6.55
Titration acid	0.08	1.12**	0.47	25.59

\* & \*\* = Significant at  $P < 0.5$  and  $P < 0.1$ , respectively; Rep = Replication; Geno = Genotypes; CV (%) = Coefficient of variation. Numbers in parenthesis indicates degree of freedom.

### 3.1.2. Mean Performance of Clones

Clones exhibited a wide range of mean values for all traits, particularly for the economically most important traits, i.e. fruit yield per hectare which ranged from 3.81 to 19.67 tons/ha, whereas the mean was 11.885 tons/ha (Table 2). The highest fruit yield was obtained from Cardaba (19.67 tons/ha) followed by Chibule Angombe (15.2 tons/ha) and Cachacko (12.78 tons/ha). Saba (12.62) also produced fruit yield comparable to the third highest yielding East Africa clone (data not shown).

The Ethiopian cooking banana clones were relatively larger in stature (size). For instance, Wondo Genet 3 had the highest mean values for pseudostem height; girth of pseudostem, number of leaves at harvest, leaf length, and width. In addition, the Ethiopian cooking banana clones had the highest mean values for bunch weight and number of finger per hand (Wondo Genet 3). On the other hand, Saba had higher mean hand weight, finger weight, finger length, finger diameter, peel weight, pulp weight and total soluble sugar.

### 3.2. Variability Components

The estimated phenotypic coefficient of variability was relatively greater than the genotypic coefficient of variation in magnitude for all the traits studied. The highest phenotypic coefficient of variation (57.22%) was observed for mean peel weight, while the lowest was computed for total soluble solid (6.67%). Similarly, the highest and the lowest genotypic coefficients of variation were obtained for the same traits i.e. mean peel weight (47.26%) and total soluble solid (1.21%) (Table 2).

The estimate of heritability in broad sense was high for mean pseudostem height (91.24%); time interval between successive leaf emergences (89.65%); pseudostem girth (85.24%); number of suckers until time of shooting (71.38%); mean finger diameter (68.34%); mean peel weight (68.23%) and mean finger weight (67.27%). Low heritability was recorded for total soluble solid (3.30%) and finger yield per hectare (12.43%). Medium heritability was computed for the remaining traits ranging from 20.02 to 59.13%. The expected genetic advance was high for mean peel weight, mean finger weight, mean pseudostem height, and mean pulp weight, while it was the lowest for total soluble solid.

### 3.3. Phenotypic and Genotypic Correlation Coefficients

The phenotypic and genotypic correlation coefficients of the 19 morpho-physicochemical traits are presented in Table 3. Fruit yield per hectare had significant and positive genetic and phenotypic correlations with all the traits except leaf length and time interval between successive leaf emergence. Number of hands had positive and significant genotypic correlations with pseudostem height, pseudostem girth, time interval between successive leaf emergence, number of leaves at harvest, leaf length, leaf breadth, mean bunch weight, mean number of finger per hand, fruit yield per hectare and titration acid. However, it had negative and significant correlations with mean finger weight, mean peel weight, mean pulp weight, mean finger length, mean finger diameter and total soluble solid. Bunch weight had a positive genotypic correlation with most of the traits except time interval between successive

leaf emergence, leaf length, total soluble sugar and titrable acid. Mean hand weight had a positive and significant genotypic correlation with pseudostem girth, leaf breadth, mean bunch weight, mean finger weight, mean peel weight, mean pulp weight, mean finger length, mean finger diameter, fruit yield per hectare and total soluble solid, while it had negative and significant associations with time interval between successive leaf emergence, leaf length and mean number of hand per bunch. Generally, considerable numbers of morpho-physicochemical traits also showed significant genotypic correlations in positive and negative directions.

Number of hands had a positive and significant phenotypic association with most of the traits except with mean pulp weight and total soluble sugar, which showed negative and significant correlations. Similarly bunch weight had positive and significant phenotypic correlations with most of the traits except negative and significant correlation with length of leaf. Mean weight of hand showed positive and significant phenotypic correlations only with leaf breadth and mean bunch

weight and negative and significant phenotypic correlation with length of leaf. Considerable number of morpho-physicochemical traits also showed significant phenotypic correlations in both directions.

### 3.4. Euclidean Distance of Cooking Banana Clones

Estimates of Euclidean distance varied from 3.52 to 9.25 with a mean and a standard deviation of 6 and 1.44, respectively. The highest distance was registered for Saba and Wendo Genet 3 (9.25) followed by Kibungo I and Wendo Genet 3 (8.68). The lowest distance was calculated for Matoke and Nijuru (3.52), followed by Wendo Genet 4 and Matoke (3.87) and Ikjmanga and Kitawira (3.90). The mean Euclidean distance result showed that the most distant clone to others was Kibungo I (6.99) followed by Saba (6.83). Cardaba (6.10) and Nijuru (5.92) were also the most distant to others next to the two. The closest clone to others was Ikjmanga (4.9) followed by Wendo Genet 3, Kitawira and Matoke (Table 4).

Table 2. Genotypic and phenotypic coefficient of variances, heritability and genetic advance in 11 cooking banana clones for 19 morpho-physicochemical traits.

Traits	Min	Max	Mean	$\sigma^2 g$	$\sigma^2 p$	GCV	PCV	H <sup>2</sup>	EGA (10%)	EGA (5%)
PSH(m)	1.422	3.49	2.017	0.28	0.31	26.42	27.66	91.24	44.41	64.60
GS(cm)	43.00	84.20	67.55	126.80	148.73	16.67	18.06	85.24	27.09	39.40
TIBSLE	5.00	10.20	7.42	2.71	3.02	22.19	23.43	89.65	36.97	53.78
NSUTS	3.40	9.00	7.02	2.79	3.92	23.82	28.20	71.38	35.42	51.53
NLH	10.00	15.00	13.00	0.67	1.96	6.30	10.76	34.29	6.49	9.45
LL(cm)	104.40	172.00	148.48	444.14	763.56	14.20	18.62	58.17	19.06	27.72
LB(cm)	52.80	67.80	59.80	14.16	35.31	6.29	9.94	40.11	7.01	10.20
MBW(kg)	5.54	11.30	8.47	4.19	10.52	24.17	38.31	39.81	26.84	39.04
MNH	5.00	7.00	5.86	0.61	1.03	13.26	17.24	59.13	17.95	26.10
MWH(g)	955.00	1422.9	1269.97	31451.50	157100.52	13.96	31.21	20.02	11.00	15.99
MNF	41.00	104.00	75.07	296.28	702.59	22.93	35.31	42.17	26.20	38.12
MFW(g)	54.98	143.00	85.35	832.06	1236.98	34.12	41.60	67.27	49.25	71.63
Mpel(g)	15.46	46.48	24.73	136.61	200.21	47.26	57.22	68.23	68.71	99.94
Mpulp(g)	39.59	87.59	59.82	337.06	598.78	30.69	40.91	56.29	40.53	58.95
MFL(cm)	9.79	15.74	12.22	2.81	5.49	13.72	19.16	51.29	17.30	25.16
MFD(cm)	2.79	4.08	3.32	0.18	0.27	12.83	15.52	68.34	18.66	27.15
MFY(t)	3.81	19.67	11.89	2.06	16.60	12.09	34.28	12.43	7.50	10.91
TSS(%)	14.28	16.44	15.12	0.01	0.11	1.21	6.67	3.30	0.39	0.56
TA(%)	1.88	4.20	2.75	0.13	0.60	13.54	28.95	21.88	11.15	16.22

Min = Minimum; Max = Maximum;  $\sigma^2 g$  = Genotypic variance;  $\sigma^2 p$  = Phenotypic variance; GCV = Genotypic coefficient of variance; H<sup>2</sup> = Heritability in broad sense; PCV = Phenotypic coefficient of variance; EGA = Expected genetic advance at 10% and 5% selection intensity; PSH(m) = Mean pseudostem height(m); GS(cm) = Pseudostem girth; TIBSLE = Time interval between successive leaf emergence; NSUTS = Number of suckers until time of shooting; NLH = Number of leaves at harvest; LL (cm) = Leaf length (cm); LB (cm) = Leaf breadth(cm); MBW(kg) = Mean bunch weight; MNH = Mean number of hand per bunch; MWH(g) = Mean hand weight; MNF = Mean number of finger per hand; MFW(g) = Mean finger weight; Mpel(g) = Mean peel weight; Mpulp(g) = Mean pulp weight; MFL(cm) = Mean finger length; MFD(cm) = Mean finger diameter; MFY(t) = Finger yield per hectare; TSS = Total soluble sugar % Brix; TA(%) = Titrable acid.

Table 3. Genotypic and phenotypic correlation coefficient above diagonal and below diagonal, respectively, for 19 morph-physicochemical traits of 11 cooking banana clones.

	PSH	GS	TIBSLE	NSUTS	NLH	LL	LB	MBW	MNH	MWH
PSH		0.76**	0.15	0.67**	0.38*	0.67**	0.56**	0.56**	0.60**	0.06
GS	0.67**		0.04	0.59**	0.53**	0.17	0.70**	0.64**	0.59**	0.40**
TIBSLE	-0.18	0.06		-0.15	-0.23	0.15	-0.15	-0.17	0.33*	-0.20*
NSUTS	0.16	0.47**	-0.15		-0.11	-0.30	0.60	0.68**	0.14	0.61
NLH	0.40**	0.39**	-0.05	-0.05		-0.02	0.29*	0.31*	0.31*	0.19
LL	0.27*	0.03	0.11	-0.23*	-0.10		-0.12	-0.35*	0.52**	-0.63**
LB	0.42**	0.37**	-0.10	0.33*	0.09	0.23*		0.79**	0.39*	0.45**
MBW	0.38**	0.40**	-0.11	0.41**	0.29*	-0.23*	0.27*		0.41**	0.57**
MNH	0.42**	0.44**	0.19	0.14	0.31*	0.21*	0.12	0.53**		-0.22*
MWH	0.00	0.15	-0.10	0.15	0.05	-0.21*	0.34*	0.33*	-0.07	
MNF	0.50**	0.45**	-0.02	0.30*	0.13	0.17	0.21*	0.60**	0.59**	0.21*
MFW	-0.09	0.22*	-0.19*	0.46**	-0.01	-0.33*	0.12	0.28*	-0.16	0.48**
MpelW	-0.10	0.17	-0.23*	0.45**	0.07	-0.34*	0.04	0.32*	0.02	0.46**
MpulpW	-0.07	0.21*	-0.14	0.40**	-0.06	-0.28*	0.15	0.22*	-0.24*	0.42**
MFL	-0.17*	0.16	-0.06	0.42**	-0.10	-0.43**	0.14	0.25*	-0.17	0.45**
MFD	-0.17*	0.23*	-0.15	0.43**	-0.08	-0.26*	0.17	0.27*	-0.16	0.55**
MFY	0.22*	0.33*	0.00	0.20*	0.19*	-0.05	0.34*	0.58**	0.50**	0.81**
TSS	-0.08	0.09	-0.04	0.16	-0.08	-0.26*	-0.04	-0.10	-0.20*	-0.05
TA	0.13	0.23*	0.13	0.06	0.07	0.10	-0.09	-0.07	0.14	0.01

	MNF	MFW	Mpel	Mpulp	MFL	MFD	MFY	TSS	TA
PSH	0.69**	-0.09	-0.10	-0.09	-0.19	-0.18	0.52**	-0.07	0.20
GS	0.65**	0.26	0.22	0.27	0.24*	0.24	0.80**	-0.03	0.35
TIBSLE	-0.02	-0.24	-0.28	-0.20	-0.08	-0.22	0.03	-0.18	0.24
NSUTS	0.46**	0.63**	0.64**	0.61**	0.67**	0.64**	0.67**	0.27*	0.09
NLH	0.18	-0.08	-0.07	-0.08	-0.12	-0.14	0.38*	-0.22	0.31
LL	0.40**	-0.45**	-0.39*	-0.47**	-0.70**	-0.42**	-0.13	-0.24*	0.32*
LB	0.48**	0.22*	0.18	0.24*	0.27*	0.17	0.69**	0.44**	-0.15
MBW	0.65**	0.40*	0.37*	0.40*	0.52**	0.34*	0.81**	0.11	-0.13
MNH	0.76**	-0.27*	-0.22*	-0.29*	-0.31*	-0.25*	0.57**	-0.27*	0.24*
MWH	0.01	0.82**	0.73**	0.86**	0.84**	0.80**	0.68**	0.36*	-0.03
MNF		-0.01	0.03	-0.04	-0.05	-0.03	0.61**	-0.31*	0.14
MFW	0.07		0.97**	0.99**	0.89**	0.98**	0.51**	0.48**	0.18
Mpel	0.12	0.85**		0.91**	0.80**	0.93**	0.48**	0.50**	0.32*
Mpulp	0.03	0.95**	0.65**		0.92**	0.98**	0.52**	0.46**	0.09
MFL	0.00	0.84**	0.67**	0.82**		0.88**	0.50**	0.35*	-0.05
MFD	0.09	0.87**	0.70**	0.85**	0.67**		0.51**	0.43**	0.11
MFY	0.53**	0.30*	0.41**	0.19*	0.28*	0.35*		0.11	0.16
TSS	-0.24*	0.28*	0.22*	0.28*	0.29*	0.18	-0.18		-0.04
TA	0.06	0.04	0.07	0.02	-0.05	0.11	0.07	-0.13	

\* & \*\*, significant at 5% and 1% probability level, respectively; Mpel(g) = Mean peel weight; Mpulp(g) = Mean pulp weight; MFL(cm) = Mean finger length; MFD(cm) = Mean finger diameter; MFY(t) = Finger yield per hectare; TSS = Total soluble sugar % Brix; TA(%) = Titrable acid; PSH(m) = Mean pseudostem height(m); GS(cm) = Pseudostem girth; TIBSLE = Time interval between successive leaf emergence; NSUTS = Number of suckers until time of shooting; NLH = Number of leaves at harvest; LL (cm) = Leaf length (cm); LB (cm) = Leaf breadth(cm); MBW(kg) = Mean bunch weight; MNH = Mean number of hand per bunch; MWH(g) = Mean hand weight; MNF = Mean number of finger per hand; MFW(g) = Mean finger weight.

Table 4. Euclidean distance of 11 cooking banana clones measured from 19 morpho-physicochemical traits

Clone	C2	C3	C4	C5	C6	C7	C8	C9	C10	C11	Mean ED
C1	4.05	4.94	8.39	8.15	8.61	6.84	5.79	8.38	8.68	6.1	6.99
C2		3.87	7.14	5.99	7.66	5.03	4.57	5.87	7.76	4.79	5.67
C3			6.6	5.64	7.57	3.52	3.96	5.57	6.84	4.93	5.34
C4				4.56	4.93	6.7	4.41	5.11	7.26	5.88	6.1
C5					5.95	6.39	4.83	4.29	7.19	4.68	5.76
C6						8.51	5.23	5.49	9.25	5.13	6.83
C7							4.98	5.54	6.13	5.6	5.92
C8								4.8	6.54	3.9	4.9
C9									7.15	5.53	5.77
C10										6.7	5.32
C11											5.32

C1 = Kibungo I; C2 = Wendo Genet 4; C3 = Matoke; C4 = Cardaba; C5 = Chibule Angombe; C6 = Saba; C7 = Nijuru; C8 = Ikjmanga; C9 = Cachacko; C10 = Wendo Genet 3; C11 = Kitawira, Mean ED= Mean Euclidean distance.

The dendrograms from UPGMA cluster analysis based on the Euclidean distance (ED) matrix is presented in Fig. 1. Clustering resulted in the formation of two groups, of which one cluster comprised a single clone viz., Wendo Genet 3, while the rest of the 10 cooking banana clones formed one big group. Within this big group, two sub-groups were formed, the first subgroup consisted of Kibungo I, Wendo Genet 4, Matoke, Nijuru, Ikjmanga and Kitawira, while the second subgroup comprised Cardaba, Chibule Angombe, Cachacko and Saba. Clones in the first sub group formed three distinct pairings, namely, Kibungo I and Wendo Genet 4, Matoke and Nijuru, and Ikjmanga and Kitawira whereas in the second sub group only Chibule Angombe and Cachacko were grouped together, while Saba and Cardaba stood solitary.

Wendo Genet 3 is characterized by long and large pseudostem height and girth; many numbers of leaves; long leaf length and broad leaf; highest bunch and hand weight; many numbers of fingers per hand, but lowest finger weight, length and diameter. The sub group consisting of six cooking banana clones was characterized by having almost similar leaf length and width, lower bunch and hand weight, lower number of fruit per hand, lower finger weight, length and diameter as well as lower peel weight. The distinguishing characteristic of this sub-group was low yield. On the other hand, the second sub-group was characterized by consisting high yielding clones. Among the four clones included in this sub-group, Cardaba and Chibule Angombe yielded 19.67 and 15.204 tons of fruit yield per hectare, respectively, which were the first and second highest yields recorded in the experiment.

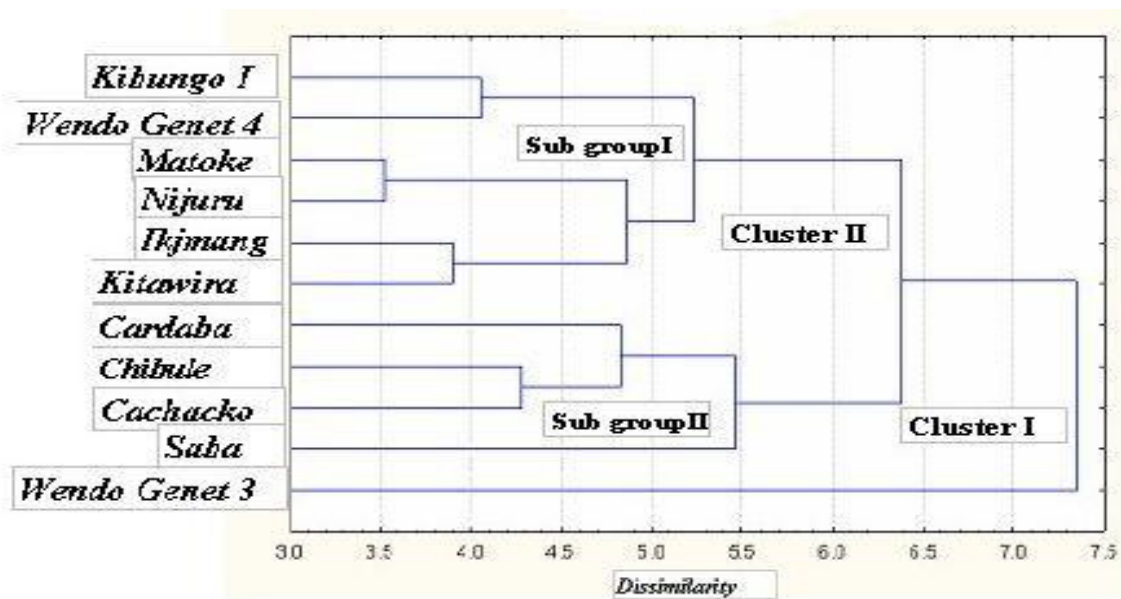


Figure 1. Dendrogram generated based on UPGMA clustering method depicting genetic relationships among 11 cooking banana clones based on 19 morpho-physicochemical traits

#### 4. Discussion

From the present study, sufficient variability was revealed by a few of the tested cultivars for all the traits except one, which would enable effective selection. The presence of variation is critical in selecting cultivars in all crops, but it is most important in sterile Bananas which develop their fruit by vegetative parthenocarpy (Pollefeys and Arnaud, 2004). The presence of variations among clones within the AAA-EA (East African cooking bananas) was previously reported (JAICAF 2010). Significant differences were also reported for banana genotypes which include cooking bananas in Nigeria for all the traits studied (Tenkouano *et al.*, 2002).

According to the category of GCV and PCV proposed by Sivasubramanian and Menon (1973), both GCV and PCV values were moderate to high for all traits studied except for number of leaf at harvest, leaf breadth and total soluble solid, which registered less than 10%. The differences between phenotypic and genotypic coefficient of variations were also very low. This indicate that the lower sensitivity of these traits to environmental modifications and the expression of these traits is more dependent on genetic factors rather than on non-genetic factors. The existence of substantial variability indicates the greater scope of improvement of the characters through selection breeding. Consistent with the results of this study, Sirisena and Senanayake (1999) also reported that estimated phenotypic coefficient of variability was relatively greater than the genotypic coefficient of variation for the traits. Tenkouano *et al.* (2002) also reported genetic differences accounted for more than 90% of the phenotypic variation for most yield traits which suggested that selection can be successfully carried out at all sites and in any crop cycle.

In this study, both heritability and expected genetic advance values were high for pseudostem length; pseudostem girth; time interval between successive leaf emergence; number of suckers until time of shooting; mean finger weight; mean peel weight and finger diameter. The result of this study was in agreement with Rodomiro (1997) who reported high heritability (>80%) for plant height and fruit size. As suggested by Panse (1957) the observed high genetic advance coupled with high heritability is an indication of more additive gene action. The heritability of a character determines the extent to which it is transmitted from one generation to the next. Heritability is a valuable tool when used in conjunction with genetic advance expectations from selection in predicting genetic gain that follows in the selection for that character (Ansari *et al.*, 2004; Singh and Upadhyay, 2013). Therefore, the improvement of the traits listed above is possible through selection since high heritability was coupled with high genetic advance percentage over mean. However, both heritability and expected genetic advance values were low for fruit yield per hectare and total soluble solid. As suggested by Singh (1990),

selection for these traits may be considerably difficult or virtually impractical due to the masking effect of environment on the genotypic effect. For the rest of the traits, either moderate heritability and low genetic advance or low heritability and moderate expected genetic advance were registered. This indicated that these traits are less amenable for selection. Tenkouano *et al.* (2002) and Ortiz (1997) reported heritability estimates for yield attributes based on data from two crop cycles were greater than those obtained with single cycle data.

The genotypic correlations were higher than the phenotypic correlations for most of the traits. In plant breeding programme, direct selection for yield as such could be misleading due to the complex relationships between grain yield and its components (Ali *et al.*, 2008). Because some of yield components are in positive correlation and others are in negative correlation, which make difficult in attaining efficiency of selection of genotypes for high yield. Improving one component usually increases or decreases the values of other component for which negative and positive correlations are observed (Ifikhar *et al.*, 2012). The present study suggests that indirect selection for bunch weight and fruit yield per hectare could be achieved by selecting for yield components such as mean weight of hand; number of fruits per hand and fruit weight where genotypic correlations among the traits were significant and greater than phenotypic correlations in magnitude. The present study result is in agreement with Tenkouano *et al.* (2002) and Sirisena and Senanayake (1999), who reported similar results, but in contrast with findings of Sirisena and Senanayake (1999).

East African cooking banana clones exhibited considerable genetic distances. The Ethiopian local banana, Wendo Genet 3 forming solitary cluster which indicates it is the most distant to other East African cooking banana clones. The formation of solitary cluster may be due to intensive natural or human selection for diverse adaptive complexes. The recurrent somatic mutations followed by human selection for their tasty fruit led to great phenotypic diversity amongst beer and cooking bananas in the highlands of East Africa (Crouch *et al.*, 1998). The presence of sufficient polymorphisms those were collectively useful in distinguishing the cultivars were reported in East African banana germplasm using RAPD analysis (Pillay *et al.*, 2001). Suryasari and Ahmad (2010) also reported the genetic diversity of 18 cooking bananas and plantains measured by RAPD and ISSR markers. Various DNA-based marker techniques were also employed in identification of specific banana cultivars and diversity study which supplied additional information. According to some reports, unlimited number of polymorphic bands were produced that are potentially used in simultaneous screening of a large number of accessions (Reddy *et al.*, 2002; Lakshmanan *et al.*, 2007; Venkatachalam *et al.*, 2007; Agoreyo *et al.*, 2008; Brown *et al.*, 2009).

## 5. Conclusion

This study showed the presence of sufficient variability among the studied few east African cooking banana clones. The results of this study have demonstrated that the genetic distance of the local Ethiopian cultivars as well as that of cooking bananas from other East African countries do not have a clear relationship with their geographical origin. However, there has been evidence of considerable genetic distances among the cultivars. This indicates the need to study a large number of local as well as other East African clones to exploit the diversity in breeding programs and to design appropriate conservation method for the germplasm

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## Effect of Urea and Lime Treatment on Chemical Composition, *In vitro* Digestibility, and *In sacco* Degradability of Sesame Straw

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**Abstract:** A study was conducted to evaluate the effect of treating sesame straw (SS) with lime and urea on the chemical composition, *in vitro* organic matter (OM) digestibility (IVOMD), and *in sacco* dry matter (DM) degradability of the straw. The treatments consisted of three levels of urea (0, 2 and 4%, w/w) and three levels of lime (0, 3 and 6%, w/w). The experiment was laid out as a completely randomized design (CRD) in a factorial arrangement and replicated three times per treatment. The results showed that urea treatment at 2 and 4% increased crude protein (CP) content of the straw by 47 and 76% from the level of 4.5% CP in the untreated SS. Urea treatment reduced the contents of neutral detergent fiber (NDF), acid detergent fiber (ADF) and cellulose in SS. Treating the straw with lime increased significantly only the level of ash due to the added calcium. *In vitro* organic matter (OM) digestibility (IVOMD), *in sacco* dry matter (DM) disappearance at most of the incubation hours, and washing loss and effective degradability of SS were increased with 4% urea treatment. However, treating the SS with lime did not improve the IVOMD and *in sacco* degradability of DM. Treating the straw with combined lime and urea also did not increase digestibility compared to the sole urea treatment. In conclusion, this study has revealed that treating sesame straw with 4% urea is the most effective method to enhance the feeding value of SS, which results from increased crude protein (CP) content and digestibility of the straw.

**Keywords:** Mineral concentrations; Organic matter digestibility; Organic matter degradability; *Sesamum indicum* L

### 1. Introduction

Different methods are widely used to improve the nutritive value of straws. These include physical, chemical, and biological methods and their combinations (Preston, 1995). Physical form of the forage influences feed utilization through its effect on mastication, microbial fermentation, rate of passage and digestion in the gastro-intestinal tract (Lu *et al.*, 2005). Thus, method of processing the feeds such as chopping has been noted to positively impact feed intake and consequently animal performance. Different alkali chemicals have been also utilized to enhance the feeding value of straws, through breaking ligno-cellulose bonds and making structural carbohydrates available for ruminal fermentation. Lime (CaO), a weak alkali, has been shown to be effective in solubilising the cell walls of straw and thus increasing its digestibility and also to supplement the ration with calcium (Ca) (Sirohi and Rai, 1998). The use of lime for alkali treatments of straws may be safer and more cost effective than the use of strong alkali like NaOH (Chaudhry, 1998). Urea ((NH<sub>2</sub>)<sub>2</sub>CO) solution in water is another commonly used straw treatment to improve the nutritive value through its possible effect on ligno-celluloses bandage and through enhancing total CP supply to the animal (Chenost, 1995). Since urea is a

solid chemical, it is also easy to handle and transport (Chenost, 1995) and urea can be obtained easily in markets of many developing countries.

Combinations of lime and urea may be attractive for straw treatment. Such a mixture would be able to combine treatment effects of both chemicals (Sirohi and Rai, 1999), with a concomitant increase in Ca and nitrogen in the treated straw. In addition, mould inhibition is an important effect of ammonia released from urea from this mixture in moist straws (Zaman *et al.*, 1994; Zaman and Owen, 1995; Pradhan *et al.*, 1997).

Sesame is a major crop widely cultivated in the western lowlands of Ethiopia. As a result, huge amounts of sesame straw are available in the area. Although the level of utilization between the sesame growing districts varies, the amount of sesame straw used as animal feed is generally low. Most of the straw is wasted mainly due to lack of awareness on the value of the resource as animal feed. Similar to other crop residues, sesame straw is generally low in nutritional value (Asma *et al.*, 2009). Therefore, efficient utilization of sesame straw necessitate designing appropriate strategies to enhance the feeding value of the straw, along with the creation of awareness about the potential of the resource as feed (Teferi *et al.*, 2013). With the increasing need to explore alternative and cost

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effective feedstuffs, research into the use of sesame straw as livestock feed is of great interest. However, limited research results have been published concerning methods to improve sesame straw as livestock feed. Therefore, this study was conducted with the objective of evaluating the effect of lime and urea treatment on the chemical composition and *in vitro/in sacco* degradability of sesame straw.

## 2. Materials and Methods

### 2.1. Description of the Study Area

The sesame straw samples were obtained from Kafta Humera district in Ethiopia. The chemical treatment of sesame straw was also done in the same district. The district is located in the north-western lowlands of Ethiopia, 960 km from the capital Addis Ababa. The area is located between 13°14' to 14°27' N latitude and 36°27' to 37°32'E longitude. The altitude of the area ranges between 550 to 1849 meters above sea level. The maximum temperature varies from 33 °C in April to 42 °C in August, while the minimum temperature ranges from 17.5 °C in July to 22.2 °C in May. The mean annual temperature is 26 °C and the average annual rainfall is 448 mm (MOA, 1998).

### 2.2. Treatments and experimental design

The treatments consisted of sesame straw treated with three levels of urea (0, 2 and 4%) and three levels of lime (0, 3 and 6%). The experiment was laid out as a completely randomized design (CRD) in a factorial arrangement and replicated three times per treatment.

### 2.3. Experimental procedure

To prepare samples for analyzing the chemical composition, *in vitro* organic matter (OM) digestibility (IVOMD) and *in sacco* dry matter (DM) degradability determination, samples of sesame straw of the cultivar *Hirbir*, which is widely cultivated in the district, were collected. The samples were collected across three locations varying in altitude from 550 and 750 meters above sea level within the same agro-ecology. The samples were taken from the field immediately after crop harvest. The straw was first hand-chopped with a knife into a size of about 4-5 cm in length. It was then taken in a one kg lots and treated with lime and urea accordingly. The chemicals were prepared in a solution to assure 0.5 litre of water in 1kg of air-dried sesame straw. After thoroughly mixing manually, the treated material was placed in double-layered polyethylene bags and sealed with the utmost care to exclude as much air as possible. The bags were then stored for 3 weeks as per the recommendations of Zaman *et al.* (1994) and Sirohi and Rai (1998). All the samples including the untreated straw were kept in a shed in an ambient temperature ranging from 22 to 33 °C. At the end of the treatment period, samples of the untreated and treated sesame straw were dried at 60 °C for 72 hours in a forced draft oven and ground and passed through a

1mm screen for determination of chemical composition and *in vitro* organic matter digestibility.

### 2.4. Chemical Analysis

After ensiling for three weeks, the samples of the untreated and treated sesame straw were taken for chemical analysis. The samples were analyzed for DM, ash, and nitrogen (N) using the procedure of AOAC (1990). Crude protein (CP) was calculated as N x 6.25. Neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) were determined following the procedure of Van Soest and Robertson (1985). For the purpose of comparison, all components except ash were calculated as percentages of the organic matter (OM) to avoid confounding with the expected increase in ash due to lime in the lime treated straw. Calcium and Magnesium concentrations were determined by the atomic absorption spectrophotometry (AAS) with a Perkin-Elmer AAS 2380 (Perkin-Elmer, 1982). Potassium and sodium concentrations were determined by flame photometry following the methods described by Chapman (1965). Phosphorus (P) concentration was determined by vanadomolybdate method and its concentration was read thereafter on the atomic absorption spectrophotometer (AOAC, 1990).

### 2.5. *In vitro* Organic Matter Digestibility

*In vitro* organic matter digestibility (IVOMD) was determined by the two-stage method of Tilley and Terry (1963). The samples were incubated for 48 hours with rumen fluid and buffer followed by another 48 hour digestion with pepsin and HCl, and the residue was ashed in a muffle furnace at 550 °C for 5 hour. Rumen fluid was obtained from rumen fistulated Boran x Holstein Frisian steers kept at Holleta Agricultural Research Station at maintenance dietary condition with diets containing hay supplemented daily with 4 kg of concentrate mixture comprising 74, 25 and 1% wheat bran, noug seed cake, and salt, respectively.

### 2.6. *In sacco* Dry Matter Degradability

All samples were ground and passed through a 2 mm screen for determination of *in sacco* DM degradability parameters. Rumen degradability of the samples were determined by incubating about 2.5 g of sample in a nylon bag (41µm pore size and 6.5 x 14 cm dimension) in three same rumen fistulated steers with similar feeding management as mentioned above. The steers were kept in individual pens. The feed samples were incubated for 0, 6, 12, 24, 48, 72 and 96 hours. Duplicate nylon bag containing samples were incubated in three rumen fistulated animals by placing the straw samples at different hours and taking them out at the same time (sequential addition). At the end of the incubation period, the bags containing the samples, including the zero hour bags were hand-washed under tap water until clear water appeared. The washed bags were then dried in an oven at 105 °C for 24 hours. The

dried bags were then taken out of the oven and allowed to cool in a desiccator, and weighed immediately.

The degradability of DM (DMD) was determined for each incubation time using the following formula;  $DMD (g\ kg^{-1}) = (DM\ in\ straw\ sample - DM\ in\ residue) / DM\ in\ straw$ . The DMD data was fitted to the exponential equation  $p = a + b(1 - e^{-ct})$  as described by Orskov and McDonald (1979), using Neway Excel program (Chen, 1997), where  $p$  is DM degradation (%) at time  $t$ . Since washing losses ( $A$ ) were higher than the estimated rapidly soluble fraction ( $a$ ), the lag time was estimated according to McDonald (1981) by fitting the model  $p = A$  for  $t \leq t_0$ ;  $p = a + b(1 - e^{-ct})$  for  $t > t_0$  and the degradation characteristics of the straws were defined as  $A$  = washing loss (readily soluble fraction);  $B = (a + b) - A$ , representing the insoluble but fermentable material;  $c$  = the rate of degradation of  $B$  and the lag phase ( $L$ ) =  $(1/c) \log_e[b/(a + b - A)]$ . The DM degradation constant values were used to estimate potential degradability ( $PD = A + B$ ) and the effective degradability ( $ED$ ) using the method of Orskov and McDonald (1979) and assuming the passage rate ( $k$ ) of  $3\ % / h$ :  $ED = a + [bc / (k + c)]$ .

### 2.7. Statistical Data Analysis

Data were subjected to analysis of variance (ANOVA) using the GLM (General Linear Model) procedure of statistical analysis system (SAS, 2002). The data were analyzed as a  $3 \times 3$  factorial with the model consisting of levels of lime, levels of urea and their interaction. When analysis of variance (ANOVA) declared significant difference among the treatment means, mean comparison was carried out by the least significant difference (LSD) at 5% level of significance.

## 3. Results and Discussion

### 3.1. Chemical Composition

The ash content of sesame straw increased ( $P < 0.001$ ) in response to the lime treatment (Table 1). On average, the ash content increased by 47 and 63% in sesame straw treated with 3 and 6% lime, respectively. The increase in the ash content of the straw is ascribable to the inherent Ca content of the lime. Likewise, urea treatment increased ( $P < 0.001$ ) the crude protein (CP) content of sesame straw by 47 and 76% for 2 and 4% urea treated sesame straw, respectively. The 76% increase in CP content of sesame straw with 4% urea treatment noted in this study was higher than the value reported by Getahun (2006) for a similar level of urea treatment of teff straw. However, the current value was lower than the one reported by Trach *et al.* (2001) for rice straw which increased by 89.7 and 181% after treatment with 2 and 4% urea, respectively. Moss *et al.* (1994) also reported a CP increment by 92.8% after treatment of wheat straw with 4% urea. Differences in the increment of CP due to ammoniation of straw among studies could be associated with variation in the loss of nitrogen (N) during treatment and during the handling of treated

straw samples until laboratory analysis. It has been noted that around one third of the urea-N applied for straw treatment is left in the straw after storage and aeration (Sundstol and Coxworth, 1984; Chenost and Kayouli, 1997). According to Chenost (1995), large increases in CP contents does not necessarily imply a good treatment effect, rather it may indicate the presence of residual urea that may result from partial ureolysis, where all urea nitrogen are not hydrolyzed to ammonia gas.

Both levels of urea treatment of sesame straw reduced the neutral detergent fiber (NDF) ( $P < 0.001$ ), acid detergent fiber (ADF) ( $P < 0.001$ ) and cellulose ( $P < 0.01$ ) contents of the straw. The hemicellulose ( $P < 0.05$ ) content increased in sesame straw treated with 4% urea but not in the one treated with 2% urea. This could be due to the relatively higher reduction in ADF as compared to NDF as a result of 4% urea treatment of sesame straw. Madrid *et al.* (1997) and Trach *et al.* (2001) noted the effectiveness of urea in solubilizing NDF and hemicellulose, but its incapability to significantly affect other cell wall components. Treatment of sesame straw with lime did not influence the contents of NDF, ADF and acid detergent lignin (ADL), and consequently hemicelluloses and cellulose levels. Contrary to the current result, Trach *et al.* (2001) reported that lime significantly reduced NDF, hemicellulose, ADF and ADL levels in straw. Rabaa ash alkali treatment of sesame straw was reported to reduce crude fiber, increase ash and CP content (Asma *et al.*, 2009). Trach *et al.* (2001) found high levels of delignification, as reflected by reduced NDF, ADF levels of rice straw, with combined use of urea and lime treatment. However, in this study, no effect of combined urea and lime was observed on fiber components.

Urea treatment of sesame straw did not have impact ( $P > 0.05$ ) on the concentration of minerals. Nevertheless, the level of Ca increased by 31 and 41% in response to the treatment with 3 and 6% lime, respectively (Table 2). All treatments, including the untreated sesame straw were rich in Ca, which is above the Ca requirement (0.20-0.82%) for all classes of sheep (NRC, 1985). The Ca content of the untreated sesame straw was higher than the values reported for untreated wheat straw (0.17%) (Nurfeta *et al.*, 2009), barley straw (0.26) (Arisoy, 1998) and teff straw (0.2%) (Getahun, 2006). , while sesame straw treated with the combination of 2% urea and 3% lime have comparable value of Ca with wheat straw (1.7%) treated in a similar combination of lime and urea (Nurfeta *et al.*, 2009).

The concentrations of P in the sesame straw subjected to all treatments were far below the recommended P requirement of 0.16-0.38% for sheep (NRC, 1985). The P concentrations of the treated and untreated sesame straw reported in this study were much lower than the concentrations reported for wheat straw (0.1%) (Nurfeta *et al.*, 2009) and barley straw (0.037%) (Arisoy, 1998) and teff straw (0.1%)

(Getahun, 2006). Higher concentrations of Ca and lower concentrations of P resulted in higher Ca to P ratio, with the highest ratio of Ca to P ranging from 207:1 in the untreated sesame straw to 296:1 in sesame straw treated with a mixture of 6% lime and 4% urea (Table 2). The Ca to P ratios reported in this study were far higher than the recommended ratios of 1:1 to 2:1 (McDonald *et al.*, 2002). It has been indicated that P concentrations in tropical grasses and straws are extremely low. Therefore, in areas where ruminants are dependent on sesame straw and grazing, provision of forage legumes or other sources of P is crucial in alleviating P deficiency and to maintain the proper Ca:P ratio (Kebreab *et al.*, 2005).

All treatments had a K concentration of about 0.01%, which is lower than the one reported by Nurfeta *et al.* (2009) for wheat straw (1.55 and 1.62%) and the 1.7% noted for barley straw (Arisoy, 1998). The K concentration in sesame straw is below the 0.5-0.8% level of requirement indicated for sheep (NRC, 1985). Magnesium concentration in sesame straw ranged from 0.122% to 0.15%. Thus, sesame straw having a Mg concentration within the recommended level of 0.12–0.18% can be considered a good source

of Mg for sheep (NRC, 1985). The Na concentration of sesame straw was low in all treatments, lower than the 1.5% reported for wheat and barley straws (Arisoy, 1998; Nurfeta *et al.*, 2009). In general, the slightly higher Na concentration in lime treated sesame straw than in the untreated straw indicates the presence of some impurities in the lime used to treat the straw.

### 3.2. *In vitro* Organic Matter Digestibility

*In vitro* OM digestibility of sesame straw was significantly affected by the interaction of urea and lime treatments ( $P < 0.01$ ). Significant differences were observed for the levels of urea treatment at 0 level of lime. Accordingly, the value for the 4% urea treated sesame straw was higher than that of the 0 and 2% urea treatments. Other means were similar ( $P > 0.05$ ) among treatments. *In vitro* OM digestibility of the sesame straw increased by 33% in response to treating it with a 4% urea solution. Lime treatment did not show any effect on the IVOMD of sesame straw. Hart and Wanapat (1992) noted a 40% increase in the IVOMD of 5% urea treated rice straw as compared to the untreated straw, the value of which is comparable with the one noted in the present study.

Table 1. Chemical composition and *in vitro* organic matter digestibility of sesame straw as affected by treatment with lime and urea

				% OM							
Factor	Level (%)	DM (%)	Ash (%DM)	CP	NDF	ADF	ADL	Hemicellulose	Cellulose	IVOMD	
Urea	0	91.0	9.82	4.47 <sup>c</sup>	71.9 <sup>a</sup>	64.3 <sup>a</sup>	11.3	7.66 <sup>b</sup>	53.0 <sup>a</sup>	32.8 <sup>b</sup>	
	2	90.3	10.4	6.59 <sup>b</sup>	68.7 <sup>b</sup>	61.1 <sup>b</sup>	11.4	7.60 <sup>b</sup>	49.7 <sup>b</sup>	36.5 <sup>b</sup>	
	4	91.1	11.0	7.89 <sup>a</sup>	68.3 <sup>b</sup>	59.2 <sup>c</sup>	10.4	9.06 <sup>a</sup>	48.8 <sup>b</sup>	43.5 <sup>a</sup>	
	SEM	0.402	0.407	0.366	0.514	0.415	0.814	0.366	0.866	1.79	
	Significance	Ns	Ns	***	***	***	Ns	*	**	***	
Lime	0	91.0	7.60 <sup>b</sup>	6.55	70.3	61.7	11.5	8.57	50.3	35.9	
	3	91.4	11.2 <sup>a</sup>	6.02	69.1	61.5	10.3	7.61	51.2	34.4	
	6	90.3	12.4 <sup>a</sup>	6.37	69.5	61.4	11.3	8.13	50.1	33.8	
	SEM	0.402	0.407	0.366	0.514	0.415	0.814	0.366	0.866	1.79	
	Significance	Ns	***	Ns	Ns	Ns	Ns	Ns	Ns	Ns	
Interaction	Lime	Urea									
	0	0	91.0	6.24	4.46	74.7 <sup>a</sup>	64.6	12.0	10.1 <sup>ab</sup>	52.5	33.1 <sup>c</sup>
	0	2	90.3	7.47	7.09	69.0 <sup>bc</sup>	61.6	10.8	7.39 <sup>bc</sup>	50.8	37.5 <sup>bc</sup>
	0	4	91.1	9.12	8.12	67.3 <sup>c</sup>	59.0	11.6	8.22 <sup>bc</sup>	47.4	43.8 <sup>a</sup>
	3	0	91.4	11.4	4.06	70.5 <sup>b</sup>	64.1	10.6	6.35 <sup>c</sup>	53.5	37.8 <sup>bc</sup>
	3	2	91.2	10.6	6.04	69.3 <sup>bc</sup>	61.2	10.8	8.06 <sup>bc</sup>	50.5	41.7 <sup>ab</sup>
	3	4	91.0	11.2	7.98	67.5 <sup>c</sup>	59.0	9.49	8.43 <sup>b</sup>	49.5	42.5 <sup>ab</sup>
	6	0	90.3	11.8	4.89	70.6 <sup>b</sup>	64.1	11.2	6.51 <sup>c</sup>	52.9	37.3 <sup>bc</sup>
	6	2	91.6	13.1	6.65	67.9 <sup>bc</sup>	60.6	12.7	7.34 <sup>bc</sup>	47.9	41.9 <sup>ab</sup>
	6	4	91.3	12.7	7.56	70.1 <sup>b</sup>	59.6	10.2	10.5 <sup>a</sup>	49.4	42.3 <sup>ab</sup>
	SEM		0.438	0.706	0.633	0.890	0.719	1.41	0.634	1.49	1.73
	Significance		Ns	Ns	Ns	**	Ns	Ns	**	Ns	**

<sup>a-c</sup>Means with different superscript letters in a column within a category differ at 5% level of significance; \* =  $P < 0.05$ ; \*\* =  $P < 0.01$ ; \*\*\* =  $P < 0.001$ ; Ns = Non-significant; SEM = Standard error of the mean; DM = Dry matter; OM = Organic matter; CP = Crude protein; NDF = Neutral detergent fiber; ADF = Acid detergent fiber; ADL = Acid detergent lignin; IVOMD = *In vitro* organic matter digestibility;

Table 2. Mineral concentrations of sesame straw as affected by treatment with lime and urea

Factor	Level (%)	DM (%)	% DM					
			Ca	P	K	Mg	Na	
Urea	0	91.0	1.38	0.00533	0.0124	0.120	0.0050	
	2	90.3	1.44	0.00580	0.0109	0.129	0.0047	
	4	91.1	1.50	0.00594	0.0113	0.127	0.0047	
	SEM	0.412	0.041	0.0004	0.0005	0.0124	0.00009	
	Significance	Ns	Ns	Ns	Ns	Ns	Ns	
Lime	0	91.2	1.16 <sup>b</sup>	0.00579	0.0112	0.122	0.0046 <sup>b</sup>	
	3	91.0	1.52 <sup>a</sup>	0.00566	0.0114	0.135	0.0047 <sup>ab</sup>	
	6	91.0	1.64 <sup>a</sup>	0.00563	0.0121	0.144	0.0050 <sup>a</sup>	
	SEM	0.412	0.041	0.0004	0.0005	0.0124	0.00009	
	Significance	Ns	***	Ns	Ns	Ns	*	
Interaction	Lime	Urea						
	0	0	90.95	1.02 <sup>d</sup>	0.00492	0.0117	0.122	0.00443
	0	2	90.31	1.15 <sup>cd</sup>	0.00593	0.0106	0.130	0.00465
	0	4	91.05	1.32 <sup>bc</sup>	0.00648	0.0111	0.126	0.00463
	3	0	91.44	1.52 <sup>a</sup>	0.00583	0.0117	0.136	0.00467
	3	2	91.20	1.40 <sup>b</sup>	0.00545	0.0112	0.128	0.00467
	3	4	91.02	1.52 <sup>a</sup>	0.00565	0.0113	0.132	0.00483
	6	0	90.30	1.54 <sup>a</sup>	0.00520	0.0137	0.150	0.00463
	6	2	91.55	1.73 <sup>a</sup>	0.00603	0.0110	0.147	0.00503
	6	4	91.25	1.67 <sup>a</sup>	0.00565	0.0114	0.140	0.00513
	SEM		0.438	0.0710	0.000730	0.000945	0.015	0.000154
	Significance		Ns	Ns	Ns	Ns	Ns	Ns

<sup>a,b</sup>Means with different superscript letters in a column within a category differ at 5% level of significance; \*  $P < 0.05$ ; \*\*\*  $P < 0.001$ ; Ns = Non significant SEM = standard error of the mean; DM = dry matter; Ca = calcium; P = phosphorus; K = potassium; Mg = magnesium; Na = sodium

Changes in the *in vitro* Organic Matter Digestibility (IVOMD) associated with urea and lime treatment of sesame straw observed in the current study is consistent with changes in cell wall fiber characteristics. It is well established that alkali treatment modifies straw cell wall fibers and induces partial cleavage of bonds between lignin and structural carbohydrates, thereby making the rumen more conducive for cellulolytic microbes to colonize and multiply in it, and enhance microbial digestion of the ingested straw (Cheng and Hungate, 1976; Silva and Orskov, 1988).

In the current study, IVOMD was responsive to urea treatment but not to lime treatment. Pradhan *et al.* (1997) showed higher *in vitro* dry matter digestibility (IVDMD) of ensiled rice straw with 4% or 6%  $\text{Ca}(\text{OH})_2$  than with 4% or 6% urea. The differences in *in vitro* digestibility in response to alkaline treatment of straws could be associated with genetic differences among the different straws. Wanapat *et al.* (1996) noted that the capacity of forages to react to alkaline treatment depends upon their botanical family, species and varietal differences. This capacity can essentially be linked to the nature of the phenolic acid/lignin linkages and to ether or ester-linked forms (Chenost, 1995).

### 3.3. *In sacco* Dry Matter Degradability Parameters

The *in sacco* DM disappearance of sesame straw at different incubation hours except at 0 and 24 hours was significantly affected ( $P < 0.05$ ) by the interaction of

urea and lime treatment (Table 3). Generally, urea treatment at 4% level improved *in sacco* DM disappearance at most of the incubation hours. Urea treatment at 4% level improved the washing loss (A) and the ED of sesame straw. This is consistent with the IVOMD observed in this study. However, lime treatment did not improve the *in sacco* DM degradability parameters. Instead, reduction in DM degradability in most incubation hours and reduction in degradation parameters with lime treatment of sesame straw was noted in the current result, the reason of which is not apparent. This suggests that lime treatment is an inefficient method to improve the digestibility of sesame straw. The slowly degradable fraction, (B), PD, rate of degradation (c) and lagtime were unaffected by urea treatment, lime treatment and their interaction ( $P > 0.05$ ).

According to Osuji *et al.* (1993), the nylon-bag technique provides a means of ranking feeds according to the rate (c) and extent of degradation (PD) of dry matter, organic matter, nitrogen or other nutritional parameters. However, in the present study, the insoluble but fermentable fraction (B), rate of degradation (c) and potential degradability (PD) values were not affected by urea treatment, lime treatment, and their interaction ( $P > 0.05$ ). Another study (Arisoy, 1998) reported that *in vitro* digestibility is a better technique for ranking feeds as compared to *in vivo* digestibility and *in sacco* degradability. Therefore, since

the IVOMD of sesame straw in the present study was significantly affected by the interaction of urea and lime treatment and by urea treatment, 4% urea treatment ranked as the best treatment of sesame straw to

enhance the feeding value of the straw. Generally, the result of the current study suggested that combining urea with lime did not have an added effect on improving the feeding value of sesame straw

		DM % disappearance													
Factors	Levels (%)	0	6	12	24	48	72	96	A	B	c	PD	ED	lagtime	
Urea	0	11.2	15.1 <sup>b</sup>	17.7 <sup>b</sup>	30.8	40.0	43.3 <sup>b</sup>	49.3	14.2 <sup>c</sup>	38.2	0.029	52.5	32.3 <sup>b</sup>	6.22	
	2	14.4	17.6 <sup>a</sup>	19.1 <sup>b</sup>	28.3	39.0	44.2 <sup>b</sup>	47.4	16.3 <sup>b</sup>	37.7	0.022	54.0	31.8 <sup>b</sup>	5.98	
	4	15.6	18.9 <sup>a</sup>	21.1 <sup>a</sup>	32.4	41.0	46.4 <sup>a</sup>	50.3	17.5 <sup>a</sup>	38.6	0.027	56.0	34.1 <sup>a</sup>	5.42	
	SEM	0.321	0.488	0.603	1.63	0.890	0.724	0.820	0.324	2.48	0.004	2.56	0.61	0.47	
	Significance	Ns	***	**	Ns	Ns	*	Ns	***	Ns	Ns	Ns	*	Ns	
Lime	0	15.3	19.1 <sup>a</sup>	21.5 <sup>a</sup>	33.4	42.3 <sup>a</sup>	45.8	51.2 <sup>a</sup>	17.2 <sup>a</sup>	38.1	0.028	55.3	34.5 <sup>a</sup>	4.90	
	3	14.4	16.2 <sup>b</sup>	18.2 <sup>b</sup>	29.9	38.8 <sup>b</sup>	44.7	48.3 <sup>b</sup>	15.4 <sup>b</sup>	37.8	0.026	53.1	32.1 <sup>b</sup>	6.39	
	6	14.2	16.2 <sup>b</sup>	18.2 <sup>b</sup>	28.4	38.9 <sup>b</sup>	43.3	47.5 <sup>b</sup>	15.5 <sup>b</sup>	38.6	0.024	54.0	31.6 <sup>b</sup>	6.33	
	SEM	0.321	0.488	0.603	1.63	0.890	0.724	0.820	0.324	2.48	0.004	2.56	0.61	0.47	
	Significance	Ns	***	**	Ns	*	Ns	*	**	Ns	Ns	Ns	**	Ns	
Interaction	L	U													
	0	0	11.5	15.7 <sup>b</sup>	17.9 <sup>bcd</sup>	30.1	37.6 <sup>bc</sup>	40.1 <sup>bc</sup>	50.1 <sup>ab</sup>	12.9 <sup>e</sup>	36.0	0.033	48.9	32.1 <sup>b</sup>	4.80
	0	2	15.5	15.0 <sup>b</sup>	17.0 <sup>bcd</sup>	30.5	36.9 <sup>bc</sup>	45.8 <sup>ab</sup>	46.2 <sup>bc</sup>	14.4 <sup>de</sup>	38.0	0.027	52.5	31.5 <sup>b</sup>	6.57
	0	4	22.0	23.0 <sup>a</sup>	25.8 <sup>a</sup>	38.3	43.6 <sup>a</sup>	49.5 <sup>a</sup>	53.7 <sup>a</sup>	20.8 <sup>a</sup>	40.5	0.030	61.2	37.5 <sup>a</sup>	3.67
	3	0	17.2	16.4 <sup>b</sup>	16.5 <sup>cd</sup>	29.6	37.2 <sup>bc</sup>	44.0 <sup>bc</sup>	47.1 <sup>bc</sup>	15.0 <sup>cd</sup>	36.9	0.023	51.9	31.2 <sup>b</sup>	6.37
	3	2	16.7	18.8 <sup>b</sup>	19.3 <sup>bc</sup>	29.4	42.3 <sup>ab</sup>	45.9 <sup>ab</sup>	49.1 <sup>abc</sup>	16.7 <sup>bc</sup>	38.3	0.027	55.0	33.7 <sup>b</sup>	6.23
	3	4	15.3	15.7 <sup>b</sup>	17.4 <sup>bcd</sup>	29.7	41.6 <sup>ab</sup>	44.3 <sup>bc</sup>	50.1 <sup>ab</sup>	15.3 <sup>cd</sup>	40.7	0.027	56.0	33.3 <sup>b</sup>	7.30
	6	0	14.5	15.2 <sup>b</sup>	16.9 <sup>c</sup>	26.1	32.9 <sup>c</sup>	39.4 <sup>c</sup>	43.0 <sup>c</sup>	14.4 <sup>de</sup>	36.7	0.020	51.1	27.8 <sup>c</sup>	5.47
	6	2	16.7	18.8 <sup>b</sup>	19.3 <sup>bc</sup>	29.4	42.3 <sup>ab</sup>	45.9 <sup>ab</sup>	49.1 <sup>abc</sup>	16.7 <sup>bc</sup>	38.3	0.027	55.0	33.7 <sup>b</sup>	6.23
	6	4	18.3	19.7 <sup>ab</sup>	20.2 <sup>b</sup>	29.4	41.9 <sup>ab</sup>	46.5 <sup>ab</sup>	49.6 <sup>ab</sup>	17.9 <sup>b</sup>	37.9	0.020	55.8	33.9 <sup>b</sup>	6.23
	SEM		1.21	1.40	1.13	2.43	1.74	1.69	2.20	0.581	4.44	0.008	4.56	1.03	0.856
	Significance		Ns	***	**	Ns	**	***	*	***	Ns	Ns	Ns	**	Ns

<sup>a-d</sup>Means with different superscript letters in a column within a category differ; \*  $P < 0.05$ ; \*\*  $P < 0.01$ ; \*\*\*  $P < 0.001$ ; Ns: Non-significant; SEM = standard error of the mean; L = lime; U = urea; A = readily soluble fraction; B = insoluble but fermentable fraction; c = the rate of degradation of B; PD = potential degradability; ED = effective degradability

#### 4. Conclusion

The present study has demonstrated that 4% urea treatment is more effective than the other treatment levels to enhance the feeding value of sesame straw through enhancing the digestibility of the straw. The combination of lime and urea treatment did not increase digestibility as compared to the sole urea treatment, and lime treatment was inefficient in improving the *in vitro* digestibility and *in sacco* degradability of sesame straw.

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## Management of Aflatoxigenic Fungi in Groundnut Production in Eastern Ethiopia

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**Abstract:** Aflatoxigenic fungal invasion and aflatoxin contamination of groundnut can occur during pre-harvest as well as post-harvest stages of production. Therefore, a field experiment was conducted in the 2010 cropping season with the objective of evaluating the effect of biofumigation and soil solarization on the population of *Aspergillus* spp. in treated soil, and to determine the effect of combined use of biofumigation and/or soil solarization and fungicide seed treatment on invasion of the crop by the fungus and its yield. The treatments consisted of solarization, biofumigation, solarization+ biofumigation and untreated control as a main plot and carbendazim, mancozeb, carbendazim + mancozeb fungicide seed treatment and untreated control as a sub-plot. The experiment was laid out as a split-plot design with three replications near Babile and Dire Dawa towns in eastern Ethiopia. Seeds of the Shulamit groundnut variety were used for the experiment. The results revealed that fungal population densities in the soil were reduced drastically due to soil solarization. Integration of soil solarization + biofumigation with mancozeb + carbendazim seed treatment significantly reduced infections by *A. flavus* and *A. parasiticus*, and increased seed yield by 42.1% and 70.9% over the control treatments at Babile and Dire Dawa, respectively. Soil solarization + biofumigation in combination with fungicide seed treatments increased seed yield by up to 19.4-42.1% at Babile and by 53-70.9% at Dire Dawa with decreased *A. flavus* and *A. parasiticus* seed infections. It could be concluded that seed treatment using carbendazim at the rate of 2 g kg<sup>-1</sup> seed and mancozeb + carbendazim at the rate of (1+2) g kg<sup>-1</sup> seed could be recommended as the best management option for controlling invasion of groundnut by *A. flavus* and for increasing the yield and quality of the crop in the region.

**Keywords:** Aflatoxins; *Arachis hypogaea* L; *Aspergillus flavus*; *A. niger*; *A. parasiticus*; Carbendazim; Mancozeb

### 1. Introduction

The total production area and yield of groundnut in Ethiopia in 2010/2011 cropping season were estimated at 49,603 hectares and 716,06.8 tons, respectively; the major production areas of the crop are located in Oromia (32967.8 ha), Benshangul-Gumuz (9968.73 ha), Southern Nations, Nationalities, and Peoples' Region (SNNPR) (635.04 ha), and Amhara (344.57 ha) Regional States (CSA, 2011). The national average yield of groundnut is 1.12t/ha (Alemayehu *et al.* 2014). However, diseases, pests, and other biotic and abiotic stresses constrain production and use of groundnut in the country (ICRISAT, 2000; Caliskan *et al.*, 2008). Moulding of groundnut kernels by fungi is the major problem in most groundnut producing countries partly because of the associated mycotoxins that are harmful to human and animal health.

Aflatoxins are the major mycotoxins occurring in groundnut and groundnut by-products. They are produced by two closely related fungal species, *Aspergillus flavus* and *A. parasiticus* (Yu *et al.*, 2004). *A. parasiticus* is the most common mould found in groundnut and groundnut by-products throughout the

world (ICRISAT, 2000). Aflatoxin contamination of groundnut can appear any time from pre-harvest to storage. Pre-harvest infection by *A. flavus* and subsequent aflatoxin contamination is more significant in the semi-arid tropics, especially when end-of-season drought occurs just before harvest. Aflatoxin contamination of groundnut is a serious hazard to humans, livestock, and poultry health, and is one of the most important constraints to groundnut trade. Fungal and aflatoxin contamination of groundnut not only imperils the health of humans and animals but also lowers market values of the crop. Humans and animals are exposed to aflatoxins primarily through consumption of groundnut or its products (Stoloff, 1983). Aflatoxin is a cause of hepatocarcinogenesis, often in conjunction with hepatitis B (Wild and Hall, 1998; IARC, 2002; Wild and Turner, 2002). Because of these concerns, most industrialized countries have developed a regulatory system for aflatoxins in groundnut. The strict quality regulations imposed by many developed countries have contributed to a severe decline in international groundnut trade; exports have fallen sharply in many developing countries, with

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correspondingly severe impacts on their economic development (ICRISAT, 2000).

The tolerable limit of aflatoxin was set at 15  $\mu\text{g kg}^{-1}$  by FAO and WHO and that of the European Union (EU) is limited to 4–15  $\mu\text{g kg}^{-1}$ , depending on the end use of the seed (ICRISAT, 2000). In Ethiopia, *Aspergillus flavus* has been reported to be the major fungus that limits production of the crop in most groundnut producing areas (Teklemariam *et al.*, 1986). Aflatoxin contamination of groundnut reaching 5 to 250  $\mu\text{g/kg}$  has been recorded in the Eastern Hararghe zone of Ethiopia and *A. flavus* was commonly associated with the seed (Amare *et al.*, 1995). Management of aflatoxin contamination, and *A. Flavus* and *A. parasiticus* is recommended through application of lime, farmyard manure, poultry manure, host plant resistance, chemical fumigation of the soil (ICRISAT, 2000), biological agents such as non-toxicogenic bacteria (*Bacillus* sp.) (Bottone and Peluso, 2003) and fungi (*Trichoderma harizianum*) (Papavizas *et al.*, 1985; Inglis and Kawchuk, 2002). Most of these options are not accessible or difficult to apply by smallholder farmers in developing countries. Hence, there is a need to develop methods that are affordable for adoption by smallholder farmers.

The need for an alternative to methyl bromide for controlling soil-borne plant diseases has prompted an increase in research, much of which involves manipulation and application of biological control mechanisms. Glucosinolates (GSL), found in *Brassica* species, are of interest due to the potential for using their degradation products as fumigants. The naturally occurring biofumigant gas (ITC) is produced by the mustard plants when the crop is chopped; incorporating this compound and the green material into the soil results in many benefits including improved soil structure, health and fertility, suppression of various soil-borne diseases and pests and increased soil microbial activity (Anonymous, 2008). Soil solarization has been identified as an environmentally sound, pesticide-free, low cost method for controlling a wide variety of soil-borne fungal plant pathogens and plant parasitic nematodes (Katan, 1981; Barbercheck and Broembsen, 1986). Fungicide seed treatment is also beneficial for control of seed-borne pathogens and its application before planting decreased pre-emergence and post-emergence damping off caused by the seed-borne pathogens. Seedling survival rates and plant vigour also showed an increase (Elwakil and El-Metwally, 2000). The small quantity of chemicals used make seed treatment not only cost-effective but also relatively user and environmentally friendly.

Application of biofumigation or solar radiation to achieve some degree of soil sterilization combined with fungicide seed treatments to eliminate aflatoxigenic fungi associated with the seed could overcome the major sources of inoculum thereby reducing aflatoxigenic fungi and aflatoxin contamination of groundnut (Hossein *et al.*, 2007; Rathod *et al.*, 2010). Such approaches can be easily adopted by smallholder

farmers and would not have adverse effect on the environment. The objectives of this study were to evaluate the effect of biofumigation and soil solarization on the population of *Aspergillus* spp. in treated soil, and to determine the effect of combined use of biofumigation and/or soil solarization and fungicide seed treatments on fungal invasion of groundnut and to elucidate the effect on groundnut seed yield.

## 2. Materials and Methods

### 2.1. Description of the Study Sites

Filed experiments were conducted at two research sub-stations of Haramaya University, namely Babile research sub-station, which is located at 9° 08' 40" N latitude and 42° 21' 30" E longitude (Abdi, 2004) and Dire Dawa research sub-station, which is located at 9° 31' N latitude and 41° 51' E longitude (Tamado *et al.*, 2002). Babile represents one of the major groundnut producing areas and is located in a semi-arid area. It has an altitude of 1650 meters above sea level and receives a total rainfall ranging from 507- 984 mm, averaging 671 mm per annum, with mean annual maximum and minimum temperatures of 28.1 °C and 15.5°C, respectively (Abdi, 2004). Dire Dawa research site has an altitude of 1160 meters above sea level and receives a mean annual rainfall 520 mm, and has mean maximum and minimum temperatures range from 28.1-34.6°C and 14.5-21.6 °C, respectively (Belay, 2002).

### 2.2. Experimental Materials

#### Plant Material

The groundnut variety named Shulamit, which was released by Melkawerer Research Centre in 1976, and is susceptible to *A. flavus* infection (Amare *et al.*, 1995), was used as the test crop in this study. Ethiopian cabbage (*Brassica carinata* A. Braun) was used as a biofumigant.

#### Fungicides

Carbendazim (Bavistin DF) and mancozeb (Unizeb 80 WP,) were used as fungicides.

#### Polyethylene Sheet

A transparent polyethylene sheet measuring 0.02 mm (0.001 inch) in thickness was used as a mulching material

### 2.3. Treatments and Experimental Design

The treatment consisted of solarization, biofumigation, solarization+ biofumigation and untreated control as a main plot and carbendazim, mancozeb, carbendazim + mancozeb fungicide seed treatment and untreated as sub-plot. The experiment was laid out as a split plot design with three replication. The main plot size was 15 m x 4 m, leaving 1.5 m space in between the plots. The main plot was divided into four sub plots that were 4 m long and 3 m wide (four ridges, with 75 cm distance

between the ridges), and 1m spacing was maintained between the sub-plots. Seeds were sown singly at 10 cm gap along the ridges. Fertilizers (DAP and Urea) were applied at the recommended rates of 60 kg N ha<sup>-1</sup> and 100 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> during the sowing time (2 and 4 June, 2010 at Babile and Dire Dawa, respectively).

## 2.4. Experimental Procedures

### 2.4.1. Soil Solarization

After ploughing the plots, the area to be solarized was leveled and made free from weeds, debris, or large clods. The soil was turned over by hand and raked smooth to provide an even surface. A two-meter wide transparent plastic sheet was joined to make 4 m width by using CM-adhesive before mulching. The polyethylene sheet was laid across the plots by hand, and anchored to the moist soil by burying the edges in a trench around the covered area. The plastic sheet was kept in place for 4 weeks from 5 May to 1 June at Babile and 7 May to 3 June 2010 at Dire Dawa to allow the soil to heat to the greatest depth possible. The meteorological data like rainfall, temperature, and relative humidity from May to October 2010 were collected from Jijiga National Meteorological Station (2010).

### 2.4.2. Biofumigation

Ethiopian mustard (*Brassica carinata* A. Braun) was grown between rows of groundnut as an intercrop and chopped by hand at flowering, and then 10.8 kg biomass of the mustard was incorporated to the soil by hoeing.

### 2.4.3. Fungicide seed treatment

Carbendazim (Bavistin DF) (2 g ai kg<sup>-1</sup> of seed) as systemic, mancozeb (Unizeb 80 WP) (3 g ai. kg<sup>-1</sup> of seed) as a contact fungicide and carbendazim + mancozeb (1+2 g ai. kg<sup>-1</sup> of seed) were used to treat the groundnut seeds at the manufacturers' recommended rates. Seeds were mixed with the fungicides 24 hours before sowing.

## 2.5. Data Collection

### 2.5.1. Soil Sampling and Temperature Recording

Soil samples were taken from 5 and 15 cm depths using a hand trowel. Composite soil samples were drawn from each sub plot before and after soil solarization, and just at crop harvest. Thirty g of the soil samples from 3 different positions were scooped into sterile polythene bags using a hand trowel, and then mixed to get 15 g of composite samples for each depth from each plot. The samples were used for determining propagule density of fungi with emphasis on the population density of *Aspergillus* species. Soil temperature was recorded during the period of soil solarization. Temperature was measured by inserting temperature probes of a digital thermometer (Biltema) into the soil at depths of 5 and 15 cm for both

solarized and non-solarized plots during solarization of the field.

### 2.5.2. Isolation of Fungi from Soil

For isolation of fungi with emphasis on *Aspergillus* spp., a soil dilution method was used. One g of soil sample collected as described above was suspended in 9 ml sterilized distilled water, which was subjected to further serial dilutions of 10<sup>-2</sup>, 10<sup>-3</sup> and 10<sup>-4</sup> (Saleem, 1994). Then 1 ml of each dilution was aseptically pipetted into sterilized Petri dishes, and then mixed with approximately 20 ml of culture medium. *Aspergillus flavus* selective medium (Rose Bengal Streptomycin Agar), which has a composition of 0.5 g peptone, 0.5 g K<sub>2</sub>HPO<sub>4</sub>, 0.5 g MgSO<sub>4</sub> · 7H<sub>2</sub>O, 20 g sucrose, 0.5 g yeast extract, 17 g agar, 25 mg rose bengal, 50 mg Streptomycin sulphate and 1 liter distilled water was used (Bell and Crawford, 1967). Streptomycin sulphate was added in an aqueous solution of cooled (50 °C) agar medium. Plates were then incubated at room temperature (25 °C) (Bacha *et al.*, 2007; Durowade *et al.*, 2008) and checked for fungal growth after 5-7 days, and the growing fungi on plates were counted and identified. The number of fungal colonies were used to calculate the number of fungal colony forming units in 1 g soil (cfu g<sup>-1</sup>), (*i.e.* the average number of colonies per plate was multiplied by the dilution factor) (Saleem, 1994). Cf u g<sup>-1</sup> was calculated for the aflatoxigenic species *A. flavus* and *A. parasiticus* and other *Aspergillus* spp. infecting groundnuts, namely, *A. niger* separately. The single growing colony from the plate was cultured into pure culture media. After growing the *Aspergillus* spp. in pure culture media, the above-mentioned species were identified based on their colony color, shape and microscopic structure by comparing with identification guidebooks.

### 2.5.3. Crop Data

The grain yield of groundnut was measured at maturity, from the central rows of each plot by harvesting the plants manually. After hand-shelling, seed yield per plot was measured using a sensitive balance and then adjusted to 9% moisture content, and yield per plot was converted into kg ha<sup>-1</sup>.

### 2.5.4. Assessment of Aflatoxigenic Fungal Infection of the Seeds

Samples of groundnut seeds were taken from the central rows of each subplot to assess aflatoxigenic fungal infection of the seeds. From each sample, 100 mature undamaged seed was selected and surface-sterilized by using 5% NaOCl for 3 minutes followed by rinsing the seed with sterilized distilled water. The seeds were then transferred to Czapek's plates (15 seeds per 14.5 cm diameter plate) and incubated for 1 week. Finally, the numbers of seeds infected by aflatoxigenic fungi were determined and the percentage of infection by *Aspergillus flavus*, *A. parasiticus* and other fungi calculated.

## 2.6. Data Analysis

Analysis of variance was performed for colony forming unit (cfu g<sup>-1</sup>) of each fungus, seed invasion for each fungus, seed yield. The Least Significance Difference (LSD) was used to separate means. All data analysis was conducted using the General Linear Model (GLM) procedure of SAS statistical version 9.2 software (SAS, 2009). Mean separation for significant interaction effects was carried out using GenStat version 12.1 software (GenStat, 2009).

## 3. Results and Discussion

### 3.1. Changes in Soil Temperature during Soil Solarization

At Babile, average soil temperatures of the solarized plots ranged from 41.2 - 78.6 °C and 40.3 - 76.5 °C at 5 cm and 15 cm soil depths, respectively (Figure 1). At Dire Dawa, the average soil temperature in the solarized plots ranged from 58.4 - 89.5 °C at 5 cm depth and 54.2 - 81.3 °C at 15 cm depth (Figure 2). The average temperature in the solarized plots at Babile decreased from the first to the fourth week perhaps due to the occurrence of rainfall during solarization (Figure 1 and Table 1). However, the temperature at Dire Dawa increased (Figure 2 and Table 1). In an

earlier study, the maximum temperature of soil in a field subjected to soil solarization with plastic mulch varied from 42 to 55 °C at the depth of 5 cm and from 32 to 37 °C at the depth of 45 cm, and control of soil pests was usually best in the upper 10-30 cm (Elmore *et al.*, 1997). At these soil temperatures, soil solarization controls a large variety of soil-borne pathogens such as *Verticillium dahlia*, which causes *Verticillium* wilt in many crops; *Fusarium oxysporum* that causes *Fusarium* wilt in some crops; *Phytophthora* sp. like *Phytophthora cinnamomi*, which causes *Phytophthora* root rot (Elmore *et al.*, 1997; Tjamos, 1998). Bacha *et al.* (2007) showed that soil solarization for 8 weeks reduced plant pathogenic fungi by 70 - 80% at various soil depths and 11 °C increase in soil temperature was enough to kill different type of pests. In the current study, during soil solarization for four weeks, increases in soil temperature by 20 - 46 °C were recorded at both sites. The diseases caused by *F. solani* and *S. rolfsii* were considerably suppressed by soil solarization for 3 weeks. The disease incidences from these fungi were reduced by up to 83.9% and 91.0%, respectively in response to increasing the average solarized soil temperature from 3.5 to 10.3 °C at 5 cm and from 1.2 to 6.6 °C at 15 cm depth compared with the non-solarized control (Widodo and Budiarti, 2009).

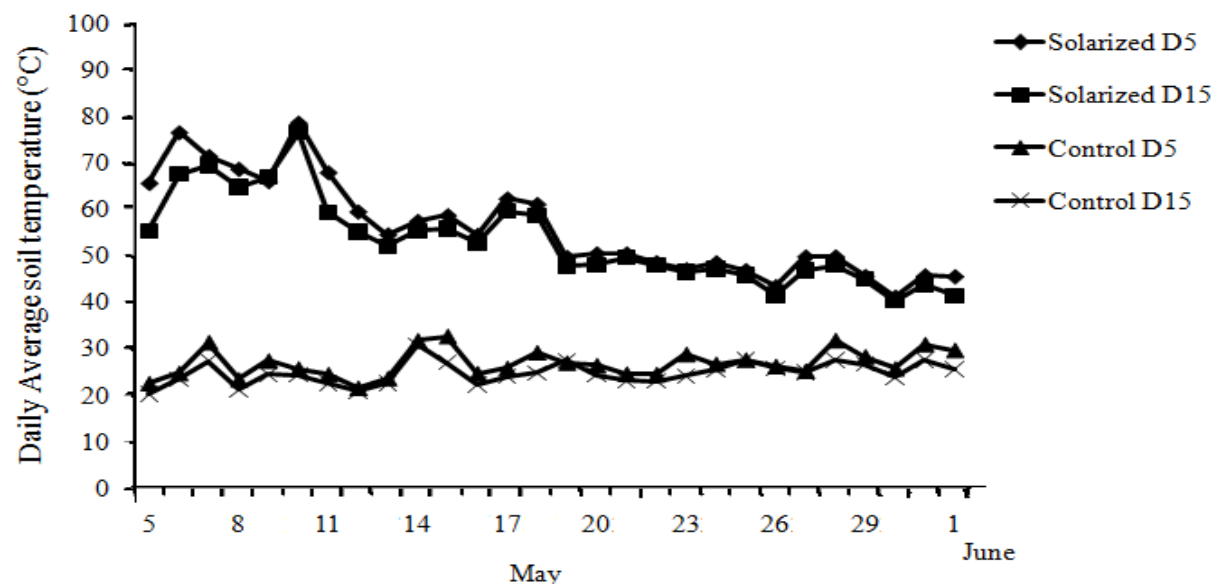


Figure 1. Daily average soil temperature (°C) during soil solarization for four weeks at Babile.

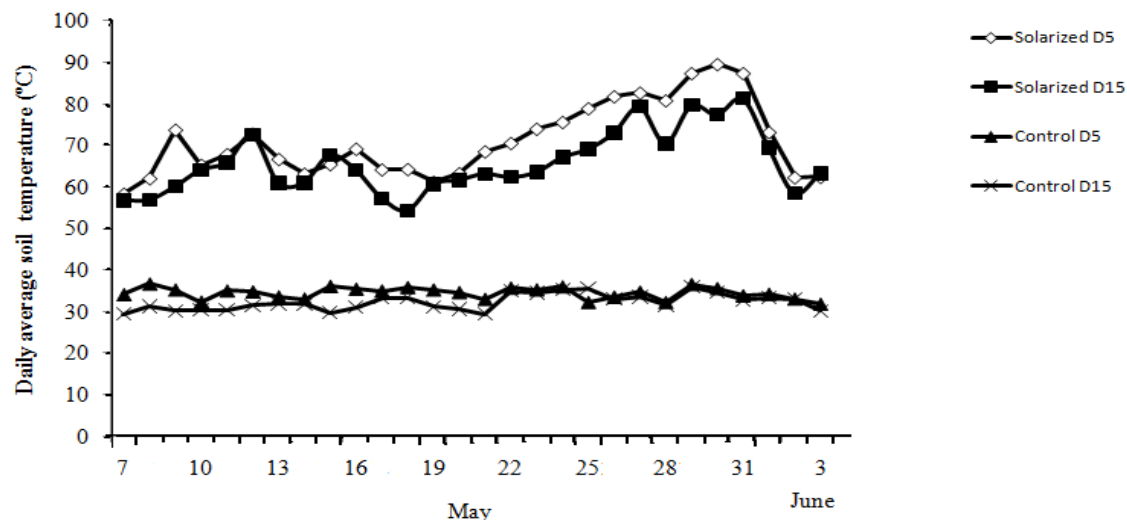


Figure 2. Daily average soil temperature (°C) during solarization for four weeks at Dire Dawa.

Table 1. Average temperature, relative humidity, total rainfall at Babile and Dire Dawa, 2010.

Month	Dire Dawa				Babile		
	Temperature (°C)		Rainfall (mm)	Relative humidity (%)	Temperature (°C)		Rainfall (mm)
	Maximum	Minimum			Maximum	Minimum	
May	35.5	22.6	75.4	36.0	27.6	15.7	197
June	36.3	23.0	15.1	35.7	28.6	14.8	92
July	33.4	21.3	119.3	56.9	24.6	14.8	59
August	33.5	21.0	194.2	49.2	24	14.5	112
September	33.3	20.6	151.8	42.8	26.7	14.8	190
October	34.9	19.0	10.7	26.7	29.1	15	56

Source: Jigjiga National Meteorological Station (2010)

### 3.2. Effect of Soil Solarization on Population Density of Soil Fungi

#### 3.2.1. *Aspergillus niger*

The data from the two main plots involving solarization, namely solarized and solarized + biofumigated, were averaged since biofumigation was applied late after planting and the two treatments were similar at this stage. Likewise, data from the non-solarized and non-solarized + biofumigated plots were averaged. Population densities of *A. niger* were reduced by  $0.17 \times 10^3$  cfu g<sup>-1</sup> at 5 cm soil depth from the initial to final colony count at Babile and by  $0.18 \times 10^3$  cfu g<sup>-1</sup> at 5 cm soil depth at Dire Dawa. Similarly, the population density of *A. niger* was decreased from the initial to final colony count at 15 cm soil depth at both sites (Table 2). Consistent with the results of this study, Bacha *et al.* (2007) reported that soil solarization was a technique that efficiently controlled *Aspergillus niger*, *Fusarium* spp., *Emereicella* spp., *Macrophomina phaseolina*, *Helminthosporium* spp. and *Verticillium* spp.

Soil solarization using a transparent plastic sheet for the four-week duration reduced *Aspergillus niger* by 77.27% and 75% at 5 cm soil depth at Babile and Dire Dawa, respectively. Soil solarization reduced plant pathogenic fungi by 70-80% at various soil depths in response to increasing the temperature by 11 °C

increase in temperature (Bacha *et al.* (2007). Sclerotia forming fungi like *Sclerotinia sclerotiorum*, *S. minor*, *Sclerotium rolfsii* and *S. cepivorum* could be controlled by soil solarization (Tjamos, 1998; Anonymous, 2008). Ramirez-Villapudua and Munnecke (1985) showed that both solar heating alone and cabbage amendments reduced soil borne populations of *Fusarium oxysporum* f. sp. *conglutinans*. In the present study, the average solarized soil temperature at 5 and 15 cm depth increased by 20-46°C compared with the non-solarized at both stations. This increased temperature reduced population density of *A. niger* by 38.89% and 46.15% at 15 cm soil depth at Babile and Dire Dawa, respectively, however its population was not reduced in non-solarized plots at both locations. Application of soil solarization reduced the population density of the pathogen from 1100 cfu/g soil to 300 after 2 weeks and reduced to 100 cfu/g soil after 4 weeks (Hossein *et al.*, 2007). Tamiatti and Valentino (2001) reported that solarization increased the soil temperature by 14.4-16.8 °C at 25 cm depth compared with the control. By these soil temperatures, solarization reduced *Verticillium* wilt severity by 89-98%.

Table 2. Colony forming unit of *A. niger*, *Aspergillus flavus* and *A. parasiticus* before soil treatment (initial count) and four weeks later (final count) at Babile and Dire Dawa towns.

Soil treatment	<i>Aspergillus niger</i> (10 <sup>3</sup> cfu g <sup>-1</sup> )				<i>Aspergillus flavus</i> (10 <sup>3</sup> cfu g <sup>-1</sup> )				<i>A. parasiticus</i> (10 <sup>3</sup> cfu g <sup>-1</sup> )			
	Initial count		Final count		Initial count		Final count		Initial count		Final count	
	D5	D15	D5	D15	D5	D15	D5	D15	D5	D15	D5	D15
<b>Babile</b>												
Solarization	0.18 <sup>a</sup>	0.13 <sup>a</sup>	0.04 <sup>b</sup>	0.06 <sup>b</sup>	0.54 <sup>a</sup>	0.44 <sup>a</sup>	0.21 <sup>b</sup>	0.32 <sup>b</sup>	0.26 <sup>a</sup>	0.24 <sup>a</sup>	0.03 <sup>b</sup>	0.07 <sup>b</sup>
Control	0.26 <sup>a</sup>	0.18 <sup>a</sup>	0.43 <sup>a</sup>	0.32 <sup>a</sup>	0.50 <sup>a</sup>	0.42 <sup>a</sup>	0.68 <sup>a</sup>	0.58 <sup>a</sup>	0.22 <sup>a</sup>	0.18 <sup>a</sup>	0.24 <sup>a</sup>	0.22 <sup>a</sup>
CV (%)	12.56	31.54	11.39	12.48	9.17	12.54	10.61	8.0	9.87	11.16	19.6	9.42
LSD (5%)	0.096	0.17	95.62	82.81	0.17	0.19	0.17	0.13	0.08	0.083	0.10	0.05
<b>Dire Dawa</b>												
Solarization	0.21 <sup>a</sup>	0.19 <sup>a</sup>	0.03 <sup>b</sup>	0.07 <sup>b</sup>	0.12 <sup>a</sup>	0.09 <sup>a</sup>	0.01 <sup>b</sup>	0.06 <sup>b</sup>	0.11 <sup>a</sup>	0.08 <sup>a</sup>	0.06 <sup>b</sup>	0.06 <sup>b</sup>
Control	0.3 <sup>a</sup>	0.26 <sup>a</sup>	0.32 <sup>a</sup>	0.26 <sup>a</sup>	0.17 <sup>a</sup>	0.13 <sup>a</sup>	0.13 <sup>a</sup>	0.12 <sup>a</sup>	0.14 <sup>a</sup>	0.11 <sup>a</sup>	0.28 <sup>a</sup>	0.29 <sup>a</sup>
CV (%)	19.2	10.61	20.25	16.89	9.4	12.25	18.84	14.41	18.36	24.96	8.16	21.6
LSD (5%)	0.17	0.083	0.13	0.10	0.06	0.048	0.05	0.04	0.082	0.083	0.05	0.13

D = soil depth at 5 and 15 cm; cfu = Colony forming unit; means within a column followed by the same letter are not significantly different at 5% level of significance; CV = Coefficient of Variation; LSD = Least Significant Difference

### 3.2.2. Effect of Soil Solarization on the Population Density of *Aspergillus Flavus* and *Aspergillus Parasiticus*

The population densities of *A. flavus* and *A. parasiticus* were significantly lower in the solarized plots than the non-solarized plots at both soil depths and locations (Table 2). This shows that soil solarization markedly reduced the number of propagules of *Aspergillus flavus* and *A. parasiticus* in the evidently as a result of increased soil temperature. Thus, at the end of solarization, the population density of *A. flavus* decreased by 61.1% and 27.3%, and that of *A. parasiticus* decreased by 88.5% and 70.8% at 5 and 15 cm soil depths, respectively at Babile. At Dire Dawa, the population density of *A. flavus* was reduced by about 92% after four weeks of soil solarization at 5 cm soil depth. The results revealed that soil solarization effectively controlled soil-borne fungi as a result of increased soil temperature to 20-46 °C in the solarized plots compared to the lower temperature in the non-solarized control plots. Th result corroborates the finding of Hossein *et al.* (2007) who reported that soil solarization was an effective non-pesticide method for controlling *A. flavus* and other soil-borne fungi in Pistachio fields.

### 3.3. Effect on of Soil Solarization, Biofumigation and Seed Treatment on the Population Density of *Aspergillus flavus*, *A. parasiticus* and *A. niger* at Crop Harvest

Colony forming units per gram of soil (cfu g<sup>-1</sup>) of *Aspergillus flavus*, *A. parasiticus* and *A. niger* showed significant difference ( $P < 0.0001$ ) at harvest in in response to the soil treatment, seed treatment and soil treatment x seed treatment at 5 and 15 cm soil depths at both locations. Biofumigation with *Brassica carinata*, coupled with fungicide seed treatment, significantly ( $P < 0.01$ ) reduced the population densities of *Aspergillus*

*flavus*, *A. parasiticus* and *A. niger* in the soil at crop harvest at both soil depths and locations (Tables 3 and 4). However, no reduction in the population density of *A. parasiticus* occurred at Dire Dawa for biofumigation coupled with mancozeb seed treatment at the soil depth of 5 cm (Table 4). Fungicide seed treatment using carbendazim and mancozeb + carbendazim significantly ( $P < 0.01$ ) reduced the population density of soil fungi at both locations. Thus, colony forming units of *A. flavus* in soil sample at harvest were significantly lower for seed treatments with carbendazim and mancozeb + carbendazim than for the untreated control plots at the depths of 5 and 15, respectively at Babile (Table 3). Similarly, significantly lower cfu g<sup>-1</sup> of *A. flavus* was obtained for carbendazim seed treatment as compared to the control treatment at the soil depths of 5 and 15 cm at Dire Dawa (Table 4).

Soil solarization integrated with carbendazim and mancozeb + carbendazim seed treatment significantly ( $P < 0.05$ ) decreased the population densities of *A. flavus*, *A. parasiticus* and *A. niger* at the two soil depths at both locations (Tables 3 and 4). Soil solarization alone significantly reduced the population densities of of *A. flavus*, *A. parasiticus* and *A. niger* compared to the control treatment at Babile. Soil solarization integrated with seed treatment with mancozeb + carbendazim and carbendazim resulted in a lower cfu g<sup>-1</sup> of *A. flavus* s compared to the untreated control treatment (at 5 cm soil depth at Babile (Table 3). Similarly, lower population densities of *A. flavus* and *A. parasiticus* were obtained in response to soil solarization combined with seed treatment with carbendazim and mancozeb + carbendazim at both soil depths compared to the untreated control treatment at Dire Dawa (Table 4). The population density of *A. flavus* was decreased in response to solarization + biofumigation significantly more than the decrease observed in response to

biofumigation + mancozeb at the two soil depths at both locations (Table 3 and 4). Similarly, lower colony forming units were recorded for lone biofumigation seed treatment than for lone mancozeb seed treatment at both soil depths at Babile (Table 3)

Integrated use of soil solarization and biofumigation using seed treatment with carbendazim and mancozeb + seed treatment with carbendazim significantly ( $P < 0.01$ ) reduced the population densities of *A. flavus*, *A. parasiticus* and *A. niger* at 5 and 15 cm soil depths at both locations. Solarization + biofumigation in combination with mancozeb markedly reduced the population density of *A. niger* at 5 cm soil depth at Babile. At Dire Dawa, the population density of *A. flavus* was lower in response to solarization + biofumigation with mancozeb + carbendazim and soil than the untreated control plots (at 5 and 15 cm soil depths, respectively (Table 4). *Aspergillus parasiticus* was not detected in soil samples from plots treated with solarization + biofumigation combined with carbendazim seed treatment. Soil solarization alone reduced the population densities of *A. flavus*, *A. parasiticus*, and *A. niger* at Babile (Table 3). Similarly, the population density of *A. flavus* in response to soil solarization alone was lower than the population density in the control treatment at 5 and 15 cm soil depths, respectively at Dire Dawa (Table 4).

Integration of soil solarization and mustard biofumigation reduced the population densities of soil-borne fungi such as *Aspergillus flavus*, *A. parasiticus* and *A. niger* at the two soil depths at both locations (Tables 3 and 4). Consistent with the results of this study, other studies revealed that soil solarization decreased soil-borne fungi such as *Phytophthora* spp., *Fusarium oxysporum* and *A. flavus* (Hossein *et al.*, 2007) and *A. niger* (Bacha *et al.*, 2007). Concordant with the results of this

study, soil solarization was used as a per-harvest method for managing aflatoxin in groundnut at soil level (ICRISAT, 2000). In the present study, it was found that the population densities of *A. flavus*, *A. parasiticus* and *A. niger* were reduced at planting in solarized soil, and at harvest in solarized + mustard-biofumigated plots at both soil depths. In agreement with the results of this study, soil solarization was found to be effective for the management of soil-borne fungi, ear rots and mycotoxins (fumonisins and aflatoxins) in fields and stored corn (Ahmad and Ghaffar, 2007).

Seed treatment using carbendazim and mancozeb + carbendazim were effective in reducing the population densities *A. flavus*, *A. parasiticus* and *A. niger* at both soil depths at both locations (Tables 3 and 4). Consistent with the results of this study, thiram, carbendazim and mancozeb were found to be inhibitory to *A. flavus* (Rathod *et al.*, 2010), and *A. flavus* and *A. niger* were controlled by 2 g kg<sup>-1</sup> seed carbendazim seed treatment (Anonymous, 2000).

In this study, integrated use of soil solarization and biofumigation with *Brassica carinata* in combination with carbendazim and along with mancozeb + carbendazim seed treatment reduced *A. flavus* and *A. parasiticus* over the untreated control plots at the two soil depths at Babile. At Dire Dawa, the combinations of soil solarization with carbendazim and solarization + biofumigation along with carbendazim markedly reduced the population densities of *A. parasiticus* up to 100% over the untreated control plots. At both locations and soil depths, population densities *A. flavus*, *A. parasiticus* and *A. niger* were reduced in response to integration of solarization with the rate of 2 g kg<sup>-1</sup> seed carbendazim and solarization with the rate of mancozeb 2 g kg<sup>-1</sup> + carbendazim 1g kg<sup>-1</sup> seed.

Table 3. Effect of solarization, biofumigation and fungicide seed treatments on colony forming units of *Aspergillus flavus*, *A. parasiticus* and *A. niger* at 5 cm and 15 cm soil depths at harvest at Babile

Soil treatment	Seed treatment	Fungal population density ( $10^3$ cfu g <sup>-1</sup> )					
		<i>A. flavus</i>		<i>A. parasiticus</i>		<i>A. niger</i>	
		D5	D15	D5	D15	D5	D15
Solarization	mancozeb	0.08	0.10	0.08	0.08	0.02	0.15
	carbendazim	0.03	0.03	0.00	0.00	0.09	0.07
	man+carb	0.01	0.01	0.00	0.02	0.00	0.01
	Control	0.10	0.12	0.12	0.10	0.23	0.11
Solar + Biofu	mancozeb	0.05	0.12	0.07	0.09	0.03	0.06
	carbendazim	0.00	0.03	0.01	0.02	0.6	0.02
	man+carb	0.00	0.04	0.01	0.00	0.04	0.04
	Control	0.07	0.13	0.11	0.16	0.11	0.09
Biofumigation	mancozeb	0.10	0.16	0.07	0.15	0.10	0.11
	carbendazim	0.01	0.06	0.04	0.07	0.10	0.04
	man+carb	0.00	0.10	0.02	0.08	0.02	0.13
	Control	0.13	0.19	0.10	0.18	0.17	0.13
Control	mancozeb	0.14	0.24	0.14	0.18	0.43	0.18
	carbendazim	0.06	0.09	0.06	0.09	0.11	0.03
	man +carb	0.01	0.11	0.04	0.09	0.06	0.06
	control	0.24	0.40	0.17	0.29	0.48	0.19
CV (%)		26.16	23.42	23.3	20.51	18.6	27.6
LSD (5%)		0.03	0.054	0.026	0.038	0.055	0.044

*Solar + Biofu* = Solarization and biofumigation; *man + carb* = Mancozeb + carbendazim; *cfu* = Colony forming unit; *D5* = Depth of soil at 5 cm; *D15* = Depth of soil at 15 cm; *CV* = Coefficient of variation; *LSD* = Least Significant Difference.

Table 4. Effect of solarization, biofumigation and fungicide seed treatments on colony forming units of *Aspergillus flavus*, *A. parasiticus* and *A. niger* in both soil depths at harvest at Dire Dawa.

Soil treatment	Seed treatment	Fungal population density ( $10^3$ cfu/g)					
		<i>A. flavus</i>		<i>A. parasiticus</i>		<i>A. niger</i>	
		D5	D15	D5	D15	D5	D15
Solarization	mancozeb	0.23	0.07	0.14	0.18	0.06	0.02
	carbendazim	0.06	0.01	0.00	0.00	0	0.05
	man+carb	0.04	0.07	0.01	0.01	0.09	0.01
	Control	0.21	0.11	0.18	0.20	0.13	0.34
Solar + Biofu	mancozeb	0.06	0.11	0.07	0.10	0.06	0.09
	carbendazim	0.06	0.07	0.00	0.00	0.01	0.07
	man+carb	0.03	0.02	0.04	0.06	0.36	0.01
	Control	0.13	0.14	0.08	0.10	0.07	0.02
Biofumigation	mancozeb	0.23	0.17	0.22	0.24	0.03	0.20
	carbendazim	0.17	0.02	0.04	0.07	0.17	0.03
	man+carb	0.06	0.09	0.10	0.11	0.06	0.00
	Control	0.11	0.21	0.22	0.16	0.68	0.89
Control	mancozeb	0.09	0.16	0.12	0.16	0.51	0.07
	carbendazim	0.08	0.04	0.06	0.08	0.04	0.07
	man +carb	0.20	0.06	0.12	0.15	0.02	0.18
	control	0.32	0.19	0.20	0.23	0.80	0.69
CV (%)		23.00	25.30	14.40	17.10	14.20	13.00
LSD (5%)		0.05	0.04	0.03	0.04	0.05	0.04

*Solar + Biofu* = solarization and biofumigation; *man + carb* = Mancozeb + carbendazim; *cfu* = Colony forming unit; *D5* = Depth of soil at 5 cm; *D15* = Depth of soil at 15 cm = Depth of soil at 5 and 15 cm; *CV* = Coefficient of variation; *LSD* = Least Significant Difference

### 3.4. Effect of Soil Solarization, Biofumigation, and Seed Treatments on Yield

Significant variations were observed on groundnut seed yield among the soil treatments ( $P < 0.05$ ), seed treatments and interaction of soil treatments x seed treatments ( $P < 0.01$ ) at both locations (Table 5).

At Babile, the highest groundnut seed yield was obtained in response to soil solarization + biofumigation and seed treatment with mancozeb + carbendazim. The seed yields of the crop obtained in response to all other treatment combinations were significantly lower than this yield at this site. The second highest groundnut seed yield at babile was obtained in response to solarization + seed treatment with mancozeb + carbendazim. The lowest seed yields were obtained from the control plots as well as from plots that were treated with sole biofumigation and sole solarization and biofumigation, which were in statistical parity. However, at Babile, solarization alone resulted in significantly higher seed yield than the seed yield obtained from the control plots as well as those attained in response to sole biofumigation and sole solarization and biofumigation. Thus, the groundnut seed yield obtained in response to solarization + biofumigation and seed treatment with mancozeb + carbendazim at Babile exceeded the seed yield obtained from the untreated plots by about 73%. Similarly, this seed yield exceeded the second highest yield at the location, which was obtained from plots treated with solarization + seed treatment with mancozeb + carbendazim by about 12%. The highest seed yield at Babile also exceeded the seed yield obtained because of lone solarization by about 35% at Babile (Table 5).

At Dire Dawa, the highest seed yields were obtained from plots subjected to soil solarization + biofumigation and seed treatment with mancozeb + carbendazim as well as from plots subjected to sole soil solarization. The yields obtained from these two treatments were in statistical parity at Dire Dawa, and were almost three times higher than the seed yields obtained from the same treatments at Babile. The second highest seed yields at Dire Dawa were obtained from plots subjected to soil solarization + seed treatment with mancozeb, soil solarization + seed treatment with mancozeb and carbendazim, soil solarization as well as from plots subjected to biofumigation, and biofumigation + seed treatment with mancozeb and carbendazim. The lowest seed yields at Dire Dawa were obtained from the control plots, control + mancozeb, biofumigation and control, soil solarization and carbendazim. Thus, at Dire Dawa, the highest seed yields obtained from the treatment with soil solarization + biofumigation and seed treatment with mancozeb + carbendazim and from sole soil solarization exceed the yield obtained from the control treatment plot by about 3.4-fold and 3-fold, in the order mentioned here (Table 5)

The lower groundnut seed yield obtained at Babile could be attributed to other groundnut diseases (rust, leaf spot and root rot/wilt) as compared to Dire Dawa.

Integrated use of soil solarization and/or biofumigation with fungicide seed treatments increased yield at both stations as compared to the untreated control plots. Soil solarization and/or mustard biofumigation increased yield per hectare at both locations. Plants often grow faster and produce yields of increased quantity and quality (size and appearance) when grown in solarized compared to non-treated soil (Elmore *et al.*, 1997; Anonymous, 2008). Corroborating the results of this study, soil biofumigation with *Brassicas* combined with soil solarization showed a high potential in the control of pathogens of the soil (Romero *et al.*, 2006) and increased crop productivity (Barrau *et al.*, 2005, 2006). Biofumigation with *Brassica carinata* improved soil structure, health, fertility, and this consequently increased yield and yield components (Anonymous, 2008). Soil biofumigation with *B. carinata* + soil solarization reduced *Phytophthora cactorum* in soil and increased strawberry yield and fruit weight (Barrau *et al.*, 2006, 2009).

Table 5. Effect of solarization, biofumigation and fungicide seed treatments on yield groundnut Shulamit variety at Babile and Dire Dawa.

Soil treatment	Seed treatment	Yield (kg ha <sup>-1</sup> ) Babile	Yield (kg ha <sup>-1</sup> ) Dire Dawa
Solarization	mancozeb	560.7	1415.6
	carbendazim	449.4	743.6
	man+carb	655.2	1500.9
	Control	542.4	1737.7
Solar +Biofu	mancozeb	579.3	1231.3
	carbendazim	526.3	1276.4
	man+carb	732.6	1988.7
	Control	457.3	1501.4
Biofumigation	mancozeb	501.4	1067.5
	carbendazim	515.7	893.2
	man+carb	585.6	1327.7
	Control	460.4	805.7
Control	mancozeb	581.2	634.5
	carbendazim	478.4	1077.3
	man+carb	501.8	1005.6
	Control	424.0	579.2
CV (%)		8.2	11.2
LSD (5%)		72.07	405.2

*Solar+Biofu* = Solarization and biofumigation; *man + carb* = Mancozeb + carbendazim; *SC* = Stand count; *CV* = Coefficient of variation; *LSD* = Least Significant Difference

Sole soil solarization increased the groundnut seed yield at both locations. However, the increments were profoundly high for Dire Dawa. This may be attributed to the inherent high temperature in the area, which becomes even more intense under solarization, having much more negative effects on pathogens, thereby suppressing their growth. This result confirms the finding Hartzet *et al.* (1993) who reported that soil solarization alone increased strawberry yield by 12% compared to the non-solarized plots. Similarly, Tamietti and Valentino (2001) reported that eggplant yield was consistently higher in the solarized plots than the non-solarized plots. Consistent with this result, Widodo and Budiarti (2009) showed that soil solarization for 1, 2 and 3 weeks enhanced peanut yields significantly.

### 3.5. Fungal Invasion of Groundnut Seed

The interaction of soil treatment x seed treatment caused significant variations ( $P < 0.05$ ) on the percentage of seed invasion by *A. niger* and *A. parasiticus*, and a highly significant variation ( $P < 0.01$ ) on percentage of infection by *A. flavus* and *Penicillium* spp. at Babile. Similarly, percentage of *A. flavus* and *A. parasiticus* ( $P < 0.01$ ) and *A. niger* ( $P < 0.05$ ) seed invasion significantly varied in response to the interaction of soil treatment x seed treatment at Dire Dawa (Table 6).

Fungicide seed treatments combined with mustard biofumigation, soil solarization and solarization +

biofumigation significantly ( $P < 0.01$ ) reduced the percentage of seed invasion by *A. flavus* and *A. parasiticus*. However, seed treatment with mancozeb, biofumigation and solarization + biofumigation did not reduce the percentage of seed invasion by *A.* (Table 6). Percentage of *A. niger* seed invasion significantly decreased in plots subjected to soil solarization with seed treatment and biofumigation with seed treatment at Babile. At Dire Dawa, carbendazim and mancozeb + carbendazim seed treatments significantly decreased percentages seed infection by *A. niger*, *A. flavus* and *A. parasiticus*. Similarly, combination of seed treatments with that mustard biofumigation, solarization + biofumigation and solarization significantly ( $P < 0.01$ ) reduced percentage of seed invasion by *A. niger*, *A. flavus* and *A. parasiticus* at Dire Dawa (Table 6). Integrated use of soil solarization and biofumigation with mustard significantly reduced seed infection by *A. flavus* and *A. parasiticus* (Table 6). Percentage of *Penicillium* spp. seed invasion was reduced in response to solarization + biofumigation in combination with carbendazim (6.7%) and mustard biofumigation with mancozeb (7.4%), but it increased in response to biofumigation with carbendazim (31.1%) and biofumigation with mancozeb + carbendazim (30.4%) at Babile. In general, soil solarization and/or mustard biofumigation integrated with carbendazim and mancozeb seed treatment were effective in reducing groundnut seed infection by *A. flavus* and *A. parasiticus* at both locations.

Table 6. Effect of solarization, biofumigation and fungicide seed treatments on seed invasion of groundnut by *Aspergillus niger*, *A. flavus* and *A. parasiticus* at both sites and *Penicillium* spp. at Babile.

Soil treatment	Seed treatment	% Seed invasion by fungi at Babile				% Seed invasion by fungi at Dire Dawa		
		<i>A. niger</i>	<i>A. flavus</i>	<i>A. parasiticus</i>	<i>Penicillium</i> pp.	<i>A. niger</i>	<i>A. flavus</i>	<i>A. parasiticus</i>
Solarization	mancozeb	48.9	1.5	4.4	25.9	60.7	22.2	2.2
	carbendazim	52.6	0.7	0.7	20.0	41.5	22.2	5.9
	man+carb	49.6	0.7	0	21.5	43.7	23.7	2.2
	Control	62.2	3.7	4.4	20	80	23	10.4
Solar + Biofu	mancozeb	40	5.2	4.4	13.3	43.7	21.5	3.0
	carbendazim	60.7	0.0	1.5	6.7	43.0	17.8	5.2
	man+carb	39.3	4.4	0.0	17.0	44.5	18.5	3.7
	Control	49.6	5.9	4.4	21.5	73.3	36.3	10.4
Biofumigation	mancozeb	47.4	5.2	3.7	7.4	55.6	25.2	5.9
	carbendazim	20.7	3.7	0.0	31.1	34.8	22.2	3.0
	man+carb	34.8	2.2	0.0	30.4	52.6	30.4	5.9
	Control	54.1	5.9	3.0	26.7	64.4	37.8	3.7
Control	mancozeb	68.9	6.7	4.4	11.9	72.9	37.0	7.4
	carbendazim	64.4	2.2	1.5	18.5	51.9	13.3	4.4
	man +carb	48.9	2.2	0.0	21.4	54.8	23.0	5.2
	control	73.3	7.4	6.7	25.2	83.7	51.9	15.6
CV (%)		20.73	26.17	28.23	23.8	11.9	17.2	27.4
LSD (5%)		17.31	1.6	1.06	8.27	11.72	7.89	2.54

*Solar + Biofu*=solarization and biofumigation, *man + carb* = Mancozeb + carbendazim, CV = Coefficient of variation, LSD = Least Significant Difference at 5% level of significance

Fungicide seed treatments reduced seed invasion by *A. niger*, *A. flavus* and *A. parasiticus* at both locations. Seed treatments using carbendazim and mancozeb + carbendazim reduced up to 70.3% of *A. flavus* and 77.6% of seed invasion by *A. parasiticus* at Babile. At Dire Dawa, carbendazim seed treatment decreased percentages of *A. flavus* seed infection by 74.4% over the control treatment. Seed treatment with carbendazim at the rate of 3g/kg seed completely reduced the growth of *Aspergillus* spp. such as *A. flavus*, *A. niger*, *A. ochraceus* and *A. parasiticus* and drastically reduced the *Aspergillus* contamination and aflatoxin production in rice (Reddy *et al.*, 2008). Combination of mustard biofumigation with that of carbendazim and mancozeb + carbendazim seed treatments were effective in reducing percentage of seed invasion by *A. parasiticus* up to 100% over the control treatment at Babile, and 80.7% and 62.3% at Dire Dawa, respectively. 90.5% of *A. flavus* seed invasion decrement was obtained on solarization integrated with carbendazim and along with mancozeb + carbendazim seed treatments at Babile, and 57.2% and 54.3% over control at Dire Dawa, respectively.

Soil solarization alone and in integration with mancozeb seed treatment gave some degree of reduction of percentage of seed infection by *A. flavus* and *A. parasiticus* at both locations. Integration of solarization + biofumigation with carbendazim and along with mancozeb + carbendazim seed treatments was effective to reduce percentage of *A. flavus* and *A. parasiticus* seed infection at both stations (Table 6). Hence, in the case of both treatment combinations marked reductions in yield loss were achieved. Seed treatment using carbendazim is recommended as a control method for *A. niger* and *A. flavus* (Anonymous, 2000). In the current study, seed treatment with carbendazim at the rate of 2 g/kg seed gave relatively some reduction of *A. niger*, and it was effective in reducing *A. flavus* at both locations. The combination of mustard biofumigation with mancozeb, solarization + biofumigation with carbendazim and along with mancozeb, and lone mancozeb seed treatments decreased percentage of seed invasion by *Penicillium* spp. at Babile (Table 6). Generally, the maximum reductions in seed yield loss due to *A. flavus* was achieved through integrated application of soil solarization + biofumigation + seed treatment with carbendazim and carbendazim at Babile. However, at Dire Dawa, the maximum reduction in groundnut seed yield losses occurred in response to both. However, at Dire Dawa, this was achieved in response to both soil solarization + biofumigation + seed treatment with carbendazim and carbendazim, and soil solarization alone.

### 3.6. Cost Benefit Analysis

At Babile, on groundnut Shulamit variety, the maximum net benefit (8370.5 Birr ha<sup>-1</sup>) was obtained from plots of mancozeb seed treatment (6156 Birr ha<sup>-1</sup>)

as compared to the control treatment. Higher net profit was also obtained from carbendazim (6830.3 Birr ha<sup>-1</sup>) and mancozeb + carbendazim (7156.3 Birr ha<sup>-1</sup>) seed treatment with 7.2% and 8.23% marginal rate of return, correspondingly at Babile. Higher net benefit was obtained from mustard biofumigation with mancozeb + carbendazim and carbendazim seed treatments (19246) and (15793.5 Birr ha<sup>-1</sup>) with additional cost 445.0 and 141.7 Birr, respectively, over control (8463.6 Birr ha<sup>-1</sup>) at Dire Dawa. The corresponding values of marginal rate of return were 2573.4% and 5432.2%. Mustard biofumigation integrated with mancozeb + carbendazim seed treatment was relatively increased yield with higher net benefit and marginal net benefit over control at both stations. Seed treatment with carbendazim effective for *A. flavus*, *A. niger* and *A. parasiticus* at both locations and had maximum net return at Dire Dawa.

## 4. Conclusions

This study has demonstrated that seed treatment using carbendazim at the rate of 2 g kg<sup>-1</sup> seed and mancozeb + carbendazim at the rate of (1+2) g kg<sup>-1</sup> seed could be recommended as the best management option for control *A. flavus* at Babile. As a result, smallholder farmers in the respective study areas should apply these management options to improve the yield and quality of groundnut in the region.

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## System Productivity of Forage Legumes Intercropped with Maize and Performance of the Component Crops in Kombolcha, Eastern Ethiopia

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**Abstract:** The highlands of Ethiopia are characterized by shrinking cultivated areas per household and scarcity of livestock feed resulting from a rapid increase in human population and expansion of arable land. Integrating forage legumes with food crops has been shown to be a useful alternative for increasing crop productivity. However, due to small land holdings, farmers are reluctant to intercrop forage legumes with food crops for fear of compromising grain yields. Therefore, a field experiment was conducted at Kombolcha Agricultural, Technical, and Vocational Education Training College located in Eastern Hararghe Zone during the 2009 cropping season. The objective of the study was to identify suitable forage legume, its optimum seed rate and sowing methods for intercropping with maize and to evaluate the effect of intercropping on system productivity, crude protein content of the maize stover, and the forage legumes. Forage legumes, vetch and lablab were row-intercropped with maize and broadcast at 25, 50, and 75% of the recommended seed rates with the recommended maize population of 44,444 plants ha<sup>-1</sup>. Sole maize and forage legumes were used as control treatments. The experiment was laid out in randomized complete block design (RCBD) in a factorial arrangement with three replications. The results showed higher mean 1000 kernel weight of maize in maize/vetch than in maize/lablab intercropping. Intercropping significantly reduced the final stand count of maize by 3.2% and that of the legumes by 10.6% compared to the sole crops. The highest dry biomass of the forage (2485 kg ha<sup>-1</sup>) was obtained from row intercropped vetch at 50% of the recommended seed rate. The highest fodder crude protein yield (849.4 kg ha<sup>-1</sup>) was obtained from row-intercropped vetch at 50% seed rate. Intercropping significantly enhanced stover crude protein content, crude protein yield and total fodder protein yield by 20, 18 and 39%, respectively as compared to the sole cropping. In conclusion, the results indicated that row intercropped vetch at 50% seed rate was more advantageous than maize-lablab intercrop.

**Keywords:** Crude Protein; Dry Biomass Yield; Intercropping; *Lablab purpureus* L.; *Vicia villosa* R.

### 1. Introduction

Mixed farming production of food crops and livestock is commonly and simultaneously practiced in the highland and mid-altitude areas. Food crops are produced for subsistence and livestock are raised to provide mainly draft power for crop cultivation and also can be sources of organic fertilizers. Various crop residues provide a considerable quantity of dry season feed supply (Alemayehu, 2004). The highland areas of Ethiopia are under stress due to shrinking of cultivated areas per household and reduced livestock feed availability (Getahun, 2008). To increase agricultural production and productivity per unit area in the small or marginal units of farming (Chatterje and Maiti, 1984) is to use intercropping as means is one to boost productivity and intensity of the land use (Ullah *et al.*, 2007).

Intercropping of maize (*Zea mays* L.) with common bean (*Phaseolus vulgaris* L.) in highland and soybean (*Glycine max* Merr.) in lowland is a common feature of the farming systems in Ethiopia (Diriba *et al.*, 2001). However, farmers commonly practice maize/bean intercropping, despite a reduction in maize grain yield up to 25% (Chemeda, 1997); intercropping of non-

food legumes for forage production with cereals is uncommon. This is, however, useful since it provides both food and feed for smallholder farmers. Thus, farmers are likely to adopt the technology of growing non-food legumes in association with cereals particularly maize to increase fodder production with less effect on maize grain yield (Amede and Kirkby, 2004). Integration of forage legumes into maize based cropping system through intercropping is one of the interventions for optimizing the productivity of a given land use (Alemayeyu, 1997; Diriba and Lemma, 2002), which can contribute towards alleviating livestock feed shortage in a mixed farming system (Mohamed-Saleem, 1986). A forage legume has a high protein concentration, palatability, and digestibility and can be useful as a supplement to livestock feed with mature cereal crop residues that are often low in nutritive value (Humphreys, 1995).

In Eastern Haraghe Zone, livestock are greatly dependent on crop residues for feed and the farmers usually harvest fodder from thinned crop plants, weeds, and defoliated leaves (Kassa, 2003). But plants could suffer from severe competition during the early growth stages and fodder production could be at the expense

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of grain yield, and thus, competition for crop residue between livestock feeding and grain yield may occur. It is thus desirable to generate alternative technologies that enable to produce forage for livestock and enhance efficient utilization of maize residue without significant change in maize grain yields. Growing of forage legumes in association with food crops is one option to improve the feeding value of crop residues (Umunna *et al.*, 1997). Legumes like hairy vetch (*Vicia villosa* R.) and lablab (*Lablab purpureus* L.) could be integrated into the system without reducing household food production, particularly maize grain yield (Amede *et al.*, 2005). However, there is lack of sufficient information on forage legumes suitable for intercropping, with maize, and the method of sowing and optimum seed rate for intercropping with the crop. Therefore, this study was conducted to identify suitable forage legumes, optimum seed rate and method of sowing seeds of the forage legumes in a maize/legume intercrop, and to evaluate the productivity and crude protein of maize stover and forage legumes in the intercropping system

## 2. Materials and Methods

### 2.1. Site Descriptions

The study was conducted at Kombolcha Agricultural Technical, Vocational Education Training (ATVET) College (42°07' 0" E latitude; 9°25' 60" N longitude; 2110 meters above sea level) located in Eastern Hararghe Zone in the Oromia Regional State. The temperature ranges from 16 to 25°C, and the annual rainfall ranges from 600 to 900 mm (BoA, 2001). The main rainy season is between July and September. The soil of the experimental site was sandy loam and contains low organic matter. Moreover, the total nitrogen and available phosphorus of the soil were found to be medium (0.13%) and low (8.77ppm), respectively according to Landon (1991) broad rating of nitrogen and available phosphorus. The pH was 5.5.

### 2.2. Experimental Materials

Two forage legumes, vetch and lablab which are assumed to have a good potential for forage production in the area, were intercropped with maize variety A511. Vetch is creeping and an early maturing forage legume. It is preferred for it is multipurpose legume, which could be used as a favorite livestock feed and for its capacity to improve soil water holding capacity, early soil cover, high biomass productivity and high feed value (Amede and Kirkby, 2004). Lablab is an erect, short-lived perennial herbaceous crop often grown as an annual (Kikafunda, 2001). It is preferred for good forage production, nutritional quality, and palatability. Lablab is also shade-tolerant so that it is suitable for intercropping. Currently, it is one of the major leguminous forage and green manure crops. Lablab is drought hardy, and has been grown in arid and semi-arid regions (Aganga and Tshwenyane, 2003). On the basis of a review of the literature, this species appears to have the best potential as an intercrop with maize

(Kevin *et al.*, 2008). The maize variety, A511 is late maturing and 245 m tall. It physiologically matures within 145 days after emergence, and is commonly used by the farmers of the area.

### 2.3. Treatments and Experimental Design

The treatments comprised row and broadcast intercropping patterns (broadcasting forage legumes under row-planted maize) and sole component crops. Each forage legume was intercropped with maize (75 cm x 30 cm) at 25%, 50% and 75% of the recommended sole seeding rate of vetch (25 kg ha<sup>-1</sup>) and lablab (35 kg ha<sup>-1</sup>). The experiment was laid out in a randomized complete block design in a factorial arrangement of 2 x 2 x 3, and replicated three times per treatment.

Thus, the treatments were as follows:

1. 100% maize + 25% vetch in rows
2. 100% maize + 50% vetch in rows
3. 100% maize + 75% vetch in rows
4. 100% maize + 25% lablab in rows
5. 100% maize + 50% lablab in rows
6. 100% maize + 75% lablab in rows
7. 100% maize + 25% vetch broadcasted
8. 100% maize + 50% vetch broadcasted
9. 100% maize + 75% vetch broadcasted
10. 100% maize + 25% lablab broadcasted
11. 100% maize + 50% lablab broadcasted
12. 100% maize + 75% lablab broadcasted
13. Sole vetch in rows
14. Sole lablab in rows
15. Sole vetch broadcasted
16. Sole lablab broadcasted
17. Sole maize in rows

### 2.4. Experimental Procedure

Maize was planted on 25<sup>th</sup> May, 2009. Fifteen days after the emergence of maize, thinning of maize to one plant per hill was undertaken and the forage legumes were drilled in between rows of maize and broadcast as per the treatment. Seeds of the sole forage legumes were sown both in row and broadcast. The row sown forage legumes were drilled as per the recommended rates at 40 cm row spacing. The broadcasted uniformly sown on the entire plot. The plot size was 4.5 m x 3.6 m. Net plot area of 3 m x 2.4 m was used as a sampling unit. At the time of planting, all the plots received a basal application of 18 kg N and 46 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> in the form of diammonium phosphate (DAP). In addition, maize plots were top-dressed with 23 kg N ha<sup>-1</sup> at knee-height stage and 23 kg N ha<sup>-1</sup> at tasseling stage. All other agronomic practices were carried out at the appropriate time.

### 2.5. Data Collection

Plant height was measured at maturity as the distance from ground level to the point where the tassel starts branching from 5 randomly taken plants per plot using meter stick. Data on yield components and yield of the

component crops were recorded. Final stand count at harvest was taken from net plot area and converted into ha. Number of kernels ear<sup>-1</sup> was recorded from 5 randomly taken ears from the net plot area. Thousand kernel weight (g) and grain yield (kg ha<sup>-1</sup>) from the net plot areas were adjusted to 12.5 % moisture content. After harvesting stover was air-dried to a constant weight and dry biomass yield (kg ha<sup>-1</sup>) was recorded. The harvest index was calculated as the ratio of grain yield to the total aboveground biomass. At harvest, samples from the aboveground parts were taken and analyzed for crude protein contents of the forage legumes herbage and maize stover. For crude protein analysis, the maize stover was taken after the grain was harvested and aboveground biomass of the forage legumes was harvested at 50% flowering. Samples were taken and air-dried for 11 days until a constant weight was obtained. The dry samples were ground to pass through a 2 mm sieve and analyzed for crude protein. Nitrogen was determined through the Macro-Kjeldhal digestion method (AOAC, 1990). The crude protein content was determined as a product of N x 6.25 (Jackson, 1962). The intercropping advantage was assessed by calculating the land equivalent ratio (LER), an index of intercropping advantage, and a reflection of the degree of inter-specific competition or facilitation in an intercropping system (Zhang *et al.*, 2011). Partial LER was used to compare in between individual LERs (LER<sub>f</sub> and LER<sub>m</sub>), which indicated competitive effects as proposed by Mead and Willey (1980).

$$LER = \frac{Y_{mm}}{Y_{ms}} + \frac{Y_{fm}}{Y_{fs}}$$

where: Y<sub>mm</sub>= Yield per unit area of maize in mixture

Y<sub>fm</sub> = Yield per unit area of forage legume in mixture

Y<sub>ms</sub> = Yield per unit area of maize in sole

Y<sub>fs</sub>= Yield per unit area of forage legume in sole

In the assessment of crop productivity of sole cropping systems, a useful expression is mass yield (mass per unit area). However, in intercropping systems, direct comparison is difficult because products are different for the different plant species growing on one piece of land. In this case, crop productivity should be evaluated using a common unit. A widely used method is the land equivalent ratio (LER) (Beets, 1982). Therefore, for the purpose of comparing the forage legumes combinations, the same standardizing factor, was used to show which combinations are genuinely more productive. Sole lablab sown in rows was used as a standardizing factor as it gave the highest forage yield for the experimental treatments.

## 2.6. Statistical Analysis

The analysis of variance (ANOVA) and comparisons of yields of intercrops with the yields sole crops at 5% probability level was carried out using the SAS Statistical Software (SAS, 2004). The Least significant difference (LSD) test at 5% level of probability was

used to separate treatment means (Gomez and Gomez, 1984).

## 3. Results and Discussion

### 3.1. Maize

#### 3.1.1. Yield Components and Yield

Final stand count at harvest, grain and stover yields of maize were not significantly affected by the main effects of forage legumes, sowing methods, and seed rates and their interactions. However, maize stand count, 1000 kernel weight, and grain yield were significantly influenced by the cropping system (Table 1).

The number sole-cropped a maize plant was significantly ( $P < 0.05$ ) higher than that of the intercropped-maize plants. The difference amounted to about 3.3% (Table 1). Maize intercropped with vetch had a significantly higher 1000 kernel weight than the one intercropped with lablab. This difference may have most likely occurred due to the differences in the growth habits of the two legume species. In addition less light interception to the crop canopy and lower photosynthetic efficiency might have resulted in lower 1000 kernels weight of maize intercropped with lablab. Consistent with the results of this study, Abraha (2013) reported a reduction in cob length of maize intercropped with lablab due to shading and competition under intercropping condition. It is obvious that the intercropped forage legumes may have exerted additional competition with maize for growth resources, thereby negatively affecting grain formation and development. The intercropping of both legumes had no significant influence on the yield (grain and stover) of maize (Table 1). This indicated the possibility of integration of forage legumes into maize without significant effect on its yield. The current result is in agreement with, the findings of Balearic and Pathway (1981) who reported that the population density and planting arrangement had no significant effect on sorghum yield in a sorghum/pigeon pea intercropping.

The maize/forage legume intercropping significantly reduced the maize grain yield by 9.5% over sole maize cropping (Table 1). However, this yield gap was narrow, which may be attributed to the fact that the legumes were under sown fifteen days after the emergence of maize. This therefore reduces the shading effects of fast growing forage legumes and makes the maize relatively competent for growth resources. The reduction in grain yield due to intercropping may be acceptable to subsistence farmers as it was below a tolerable range (10-15 %) as assumed by Nnadi and Haque (1986). Reduced maize yield was also reported in different intercropping treatments as compared to sole cropping treatments (Kimani *et al.*, 1999). Corroborating the results of this study, Francis (1986) reported a reduction in maize yield by 31% when intercropped with climbing bean. Similarly, Mpairwe (2002) reported a 20% reduction in maize grain yield

due to maize/lablab intercropping regardless of sowing methods relative to sole stands.

Table 1. Effect of forage legumes, their seed rates and cropping systems on stand count at harvest, 1000 kernel weight, grain and stover yield, crude protein content and yield of maize.

Treatment	Crop stands count (%)	1000 kernel weight(g)	Grain yield (kg ha <sup>-1</sup> )	Stover yield (kg ha <sup>-1</sup> )	Stover crude protein content (%)	Stover crude protein yield (kg ha <sup>-1</sup> )
Forge legumes						
Vetch	97.2	298.8	4806	10939	4.20	459.4
Lablab	96.4	286.8	4608	10788	5.89	633.4
LSD (P=0.05)	-	11.29	-	-	0.22	46.70
F-test	NS	*	NS	NS	**	**
Sowing methods						
Row	96.2	289.7	4445	10585	5.17*	550.8
Broadcast	97.4	295.9	4969	11143	4.91	540.0
LSD (P=0.05)	-	-	-	-	0.22	-
F-test	NS	NS	NS	NS	*	NS
Seed rate						
25%	97.4	187.0	4563	11058	4.59	507.6
50%	96.9	298.7	4959	10928	5.01	547.5
75%	96.1	292.8	4598	10605	5.53	586.5
LSD (P=0.05)	-	-	-	-	0.27	57.21
F-test	NS	NS	NS	NS	**	*
CV%	14.25	5.57	18.07	11.88	6.26	12.39
Cropping systems						
Intercropped maize	96.8	292.8	4707	10864	5.09	553.0
Sole-cropped maize	100.0	302.5	5199	10972	4.08	449.5
LSD (P = 0.05)	3.38	9.38	425.7	-	0.22	92.63
F-test	**	*	*	NS	*	*
CV (%)	1.67	1.39	3.8	6.7	2.11	8.17

LSD = Least significant difference; CV = Coefficient of variation; NS = Not significant; \* = Significantly different at  $P < 0.05$  and \*\* = Significantly different at  $P < 0.01$

### 3.1.2. Maize Stover Crude Protein

The analysis of variance showed that all main and interaction effects of forage legume species seed rate, and sowing methods significantly ( $P < 0.05$ ) affected maize stover crude protein content. The analysis also indicated a highly ( $P < 0.01$ ) significant effect of forage legumes and their seed rates on stover crude protein yield (Table 1 and 2). Maize stover crude protein content and yield were significantly higher by 28.7 and 27.8%, respectively, for maize/lablab than for maize/vetch intercropping (Table 1). This significant increase might be due to less dry matter partitioning to grain for the maize/lablab intercropping so the N may have remained in the vegetative part of maize. This may be related with higher competition of lablab with maize for growth factors as compared to vetch that hampered the dry matter partitioning to grain. Stover crude protein yield was significantly enhanced at 75% as compared to 25 and 50% of the recommended seed rates.

The results also showed that there was a significant ( $P < 0.05$ ) difference in both stover crude protein content and yield due to the cropping systems (Table 1). Intercropping of forage legumes with maize

increased stover crude protein content and the crude protein yield of stover on average by 19.8% (4.08 vs. 5.09%) and 18.7% (449.5 vs. 553.0 kg ha<sup>-1</sup>), respectively as compared to the sole maize cropping system (Table 1). One of the important benefits of integrating forage legumes with cereals is increasing the crude protein contents of animal feed (residues) which is the major determining factor for animal productivity (Murphy and Colucci, 1999). In agreement with this result, Haque (1984) reported higher protein yields from intercropping sorghum and/or maize with lablab than sole cropping. Nnadi and Haque (1986) also reported that intercropping cereals with forage legumes can be productive in terms of biomass and protein yields. Similarly, Anil *et al.* (2000) and Dawo *et al.* (2007) also reported that legumes increased the crude protein concentration when grown in mixture with maize. Furthermore, Abdollah *et al.* (2008) also concluded that intercropping of maize/berseem (*Trifolium alexandrinum*) resulted in increased crude protein contents of the maize stover compared to the protein contents of the maize stover from the sole-cropped system. Kevin *et al.* (2008) also summarized that maize/lablab, maize/soybean and maize/cowpeas intercropping increased the crude protein concentrations of maize

stover by 44, 19-36, and 9%, respectively. Azraf-ul-Haq *et al.* (2007) reported that intercropping of forage legumes improved both the fodder yield and quality.

The results revealed that intercropping maize with forage legumes can be used to improve the nutritional quality of the maize stover in terms of crude protein content as well as crude protein yield per unit of land. The results indicated the highest and lowest crude protein contents were due to 75% lablab and 25% vetch seed rates, respectively (Table 2). There was a significant difference between vetch and lablab at each seed rate and sowing methods at 75% seed rate, but such a trend was not observed between vetch and lablab at 25% and 50% seed rates and the sowing methods at the same seed rates. Generally, row-intercropping resulted in higher crude protein concentrations in the maize stover than broadcast intercropping at both seed rates except at 50%. Mpairwe *et al.* (2002) reported that the crude protein of the fodder was not affected by planting method; but, intercropping forage legumes with cereals generally resulted in fodder with higher protein concentration than fodder from sole cereals. Concurrent with the results of this study, Haque (1984) also reported that the highest crude protein yields were obtained from treatments in which two rows of sorghum and one row of lablab were planted.

Table 2. Interaction effect of forage legumes and sowing methods with the seed rates on crude protein content (%) of stover.

Forage legumes	Seed rates		
	25%	50%	75%
Vetch	3.70	4.02	4.88
Lablab	5.47	6.00	6.18
Sowing methods			
Row	4.69	4.97	5.85
Broadcast	4.48	5.04	5.21
LSD (P=0.05)		0.39	
F-test		*	
CV (%)		6.3	

LSD = Least significant difference; CV = Coefficient of variation; \* = Significantly different at  $P < 0.05$

This study showed that, regardless of their competitive effects on maize grain yield, higher seed rates of forage legumes resulted in enhanced crude protein concentrations in the maize stover. Also, the results indicated that lablab contributed more to the improvement of maize stover crude protein content than vetch as an intercrop (Table 2). This difference might be due to the inherent differences between vetch and lablab in influencing the value of protein in the associated crop. Consistent with this suggestion, Kikafunda *et al.* (2001) concluded that, legumes vary in their rates of nitrogen fixation and the amount and the time of fixed nitrogen availability to the crop in mixtures is affected by different factors. The results

obtained in the current study are in line with the findings Mpairwe *et al.* (2002) and Berhanu (2004) who reported that increased proportion of forage legumes like lablab and vetch in intercrops increased the crude protein contents in the of main crops such as maize, sorghum and oat residues. Kevin *et al.* (2008) found out that lablab intercropped with maize had the greatest potential out of the three beans [lablab, velvet bean (*Mucuna pruriens*), scarlet runner bean (*Phaseolus coccineus*)] to for increasing protein concentration over monoculture maize.

## 3.2. Forage Legume Component

### 3.2.1. Final stand count

Analysis of variance for the stand count of the forage legumes at harvest showed a significant difference ( $P < 0.05$ ) due to forage legumes and seed rates, but sowing methods and interactions had no significant effect (Table 3). For the main effect of forage legumes, a higher stand count was recorded for vetch as compared lablab (Table 3). The variation in stand count at harvest between the forage legumes could be due to the inherent ability of each legume to compete with maize. Lablab is a robust herbaceous plant and so easily outgrows other plants in competition for plant growth factors than vetch (Andrea and Pablo, 1999).

Table 3. Effect of forage legumes, their seed rates and cropping systems on stand count at harvest, dry biomass, crude protein content and yield of forage legumes.

Treatments	Stand count (%)	Dry biomass (kg ha <sup>-1</sup> )	Crude protein content (%)	Crude protein yield (kg ha <sup>-1</sup> )
Forge legumes				
Vetch	96.5	1446	16.25	235.0
Lablab	82.2	1000	13.45	134.5
LSD (P=0.05)	9.50	182	1.41	28.79
F-test	**	**	**	**
Seed rate				
25%	98.8	877	15.67	137.4
50%	87.0	1524	15.58	237.4
75%	82.3	1269	13.27	168.4
LSD (P=0.05)	11.33	222.47	1.72	35.25
F-test	*	**	**	**
CV (%)	14.3	21.5	13.7	22.6
Cropping systems				
Intercropped				
maize	89.4	1228	14.85	182.4
Sole cropped				
maize	100.0	4809	15.11	718.4
LSD (P=0.05)	3.38	1567.76	-	204.65
F-test	*	*	NS	*
CV (%)	1.7	22.9	4.6	10.6

LSD = Least significant difference; CV = Coefficient of variation; NS= Not significant; \* = Significantly different at  $P < 0.05$  and \*\* = Significantly different at  $P < 0.01$

The final stand count was significantly reduced with increase in the seed rates (Table 3). This most likely explanation might be stiff intra- and inter-specific competition for growth resources that might have resulted in mortality of some plants, consequently lowering stand count at higher seed rates. The results also revealed that the stand count of the intercropped forage legumes at harvest was significantly reduced by 10.6% as compared to sole legumes. This could be due to the reduction in the number of plants per unit area not only due to inter- but also as a result of intra-specific competition.

### 3.2.2. Aboveground Dry Biomass Yield

Aboveground dry biomass yield was significantly ( $P < 0.05$ ) affected by forage legumes, seed rates (Table 3) and the interaction of forage legumes, sowing methods and seed rates (Table 4). Aboveground dry biomass yield was significantly reduced by 74.5% in the intercropping as compared to the sole cropping system (Table 3). In this experiment, forage legumes were sown two weeks after the emergence of maize to reduce the competitive effect on maize. Tessema (2001) also reported that the growth and yield of under sown forage legumes were lower in contrast to sole cropped forage legumes, which may possibly be due to restricted light penetration and/or competition for light where maize crop was at full vegetative stage. Furthermore, Amede *et al.* (2005) reported a high vetch biomass reduction in intercropping of two maize varieties, ACV6 and A511.

The interactions of forage legumes, sowing methods and seed rates of forage legumes showed highly significant effect on dry biomass yield (Table 4).

Table 4. The interaction effect of forage legumes, their sowing methods and seed rates on aboveground dry biomass ( $\text{kg ha}^{-1}$ ) of forage legumes.

Forage legumes	Seed rates	Sowing methods	
		Row	Broadcast
Vetch	25%	948	1096
	50%	2485	1465
	75%	1143	1541
Lablab	25%	728	735
	50%	1010	1136
	75%	1368	1023
LSD ( $P=0.05$ )		256.88	
F-test		**	
CV (%)		21.5	

LSD = Least significant difference; CV = Coefficient of variation; \*\* = Significantly different at  $P < 0.01$

The highest and the lowest aboveground dry biomass yields were obtained from row-intercrop vetch at 50% and row-intercropped lablab at 25% seed rates, respectively (Table 4). In broadcast-intercropped vetch and row-intercropped lablab, there was an increase in the aboveground dry biomass yield with the increase in

seed rates, but such a consistent trend was not observed in the row-sown vetch and the broadcast lablab. Therefore, this study showed that vetch is a promising forage legume to be integrated with maize.

In comparison to all combinations of vetch with corresponding combination of lablab, the biomass yield of vetch was superior in all cases except in the combination of vetch sown in row at 75% sowing rate. One possible explanation for the highest vetch biomass may be due to its ability to exploit growth resources efficiently with less intra- and inter-specific competition. This finding is in agreement with that of Tessema and Demekash (2001) who reported that vetch was the highest yielding forage legume species compared to the others when under-sown in maize.

### 3.2.3. Forage Legumes Crude Protein

The crude protein contents of the forage legumes were significantly ( $P < 0.05$ ) affected by main effects of forage legumes, seed rates and interactions of forage legumes with sowing methods and forage legumes with seed rates (Table 5). As indicated in Table 5, the crude protein content (dry matter basis) of intercropped vetch showed a decreasing trend with increasing seeding rates for vetch; however, such a trend was not observed for lablab. Significant reductions were observed only at 75% as compared to 25% of the recommended seed rate but such a trend was not observed due to lablab. The highest (18.4%) and the lowest (12.5%) crude protein contents were obtained from vetch at 25% and lablab at 75% seed rates, respectively. The intercropped lablab showed a significantly higher protein content (14.7%) when row-intercropped as compared to when broadcast-intercropped (12.2%). However, the broadcast-intercropped vetch had the highest protein content, but it did not differ significantly with row-intercropped vetch and row-intercropped lablab (Table 5).

Table 5. Interaction effect of forage legumes with their seed rates and sowing methods on crude protein content (%) of intercropped forage legumes.

Forage legumes	Seed rates			Methods of sowing	
	25%	50%	75%	Row	Broadcast
Vetch	18.4	16.4	14.0	16.0	16.5
Lablab	13.0	14.8	12.5	14.7	12.2
LSD ( $P=0.05$ )	2.43			1.99	
F-test	*			*	
CV (%)	13.4			13.7	

LSD = Least significant difference; CV = Coefficient of variation; \* = Significantly different at  $P < 0.05$

Total crude protein yield of forage legumes was significantly ( $P < 0.05$ ) affected by all main and interaction effects except sowing methods and the interaction of forage legumes with sowing methods (Table 6). In the overall comparison, the highest crude

protein yield (425.7 kg ha<sup>-1</sup>) was obtained from the row-intercropped vetch at 50% sowing rate and the lowest (82.0 kg ha<sup>-1</sup>) was obtained from broadcast-intercropped lablab at 25% (Table 6). The protein yield of the row-intercropped lablab significantly increased as seed rate increased from 25 to 50%. The same trend

was observed in broadcast-intercropped vetch, but a significant difference existed only between the lowest and the highest seed rates. However, such a trend was not observed for row-intercropped vetch and broadcast-intercropped lablab.

Table 6. Interaction effect of forage legumes with their sowing methods and seed rates and cropping systems on total crude protein yield of the intercropped forage legumes and fodder.

		Sowing methods			
		Forage		Fodder	
Forage legumes	Seed rates	Row	Broadcast	Row	Broadcast
Vetch	25%	172.7	197.7	588.5	615.6
	50%	425.7	223.2	849.4	685.7
	75%	139.8	242.7	705.0	702.0
Lablab	25%	114.1	82.0	846.8	663.3
	50%	157.1	154.2	729.2	786.8
	75%	178.9	119.0	831.5	807.3
LSD (P=0.05)		40.71		121.57	
F-test		**		**	
CV (%)		22.6		9.8	
Cropping systems					
Intercropping		182.4		735.4	
Sole cropping		718.4		449.5(maize)	
LSD (P=0.05)		204.65		105.82	
F-test		**		*	
CV (%)		10.6		7.9	

LSD = Least significant difference, CV = Coefficient of variation; \* = Significantly different at  $P < 0.05$  and \*\* = Significantly different at  $P < 0.01$

Crude protein (kg ha<sup>-1</sup>) of forage legumes was highly significantly ( $P < 0.01$ ) affected by intercropping (Table 6). It was reduced by 74.6% (82.4 vs 718.4 kg ha<sup>-1</sup>) in intercropping as compared to the sole cropping (Table 6) almost with the same percentage reduction with dry biomass yield (1228 vs 4809 kg ha<sup>-1</sup>) (Table 1). This was expected as the legumes plant population in the intercrop was below the optimum population of the sole.

Total crude protein yield (kg ha<sup>-1</sup>) of fodder (maize stover + forage) was significantly affected by the main effects, all interactions except the interaction effect of forage legumes with sowing methods and sowing methods with seed rates. It was also significantly affected by cropping systems (Table 6). The total crude protein yield of fodder was highest (849.4 kg ha<sup>-1</sup>) and lowest (588.5 kg ha<sup>-1</sup>) in row-intercropped vetch at 50% and 25% seed rate, respectively. The protein yield of the fodder increased as seed rate increased in both legumes for broadcast intercropping, but such a trend was not observed for row intercropping (Table 6). In general, maize/forage legumes intercropping significantly increased the total fodder (maize stover + forage) crude protein yield by 63.6% as compared to sole maize (Table 6). This could be partly due to additionally produced forage in the intercropping without significant reduction in stover yield as compared to sole maize and partly due to improved crude protein content of the stover through

intercropping (Table 1). Similarly, Diriba and Lemma (2002) reported a higher percentage contribution of forage legume component to total biomass in maize/forage legumes intercropping, which in turn resulted in a higher protein yield per unit of land. Consistent with the results of this study, Azraf-ul-Haq *et al.* (2007) also reported the possibility to improve the feed quality of cereals through intercropping with forage legumes.

### 3.3. Productivity of Maize/Forage Legumes Intercropping

Significantly higher total LER was obtained from maize/vetch (1.24) as compared to maize/lablab (1.14) intercropping (Table 7). This could be ascribed to the difference in the amount of dry biomass produced; vetch was superior in this attribute than lablab (Table 1), which was manifested by higher LER of vetch (0.27) than lablab (0.18) intercropped with maize (Table 7). Most of this variation might be associated with their genetic differences and vegetative growth habits. The higher yield of vetch over lablab might be the reason for the difference, which was observed in total land equivalent ratio. Furthermore, total LER showed an increasing trend as the seed rate of the intercropped forage legumes increased from 25% to 50% and then declined with further increase in seed rate (Table 7).

Ofori and Stern (1987) pointed out that the value of LER follow the density of the legume component.

However, it is obvious that the optimum plant density could be achieved at certain points; to this effect optimum plant density was achieved in this study at 50% of the sole seed rate of forage legumes (Table 7). Similarly, in maize/bean intercropping Tsubo *et al.*

(2003) reported that yield advantages in intercropping were 17%, 26% and 15% for low (4.3 plants/m<sup>2</sup>), medium (8.6 plants/m<sup>2</sup>) and high (13.0 plants/m<sup>2</sup>) plant densities, respectively.

Table 7. Partial and total LERs of component crops.

Treatments	Partial LERs		Total LER Legumes
	LERf	LERm	
M+25% Vetch in row	0.17	0.99	1.16
M+50% Vetch in row	0.47	0.99	1.46
M+75% Vetch in row	0.21	0.96	1.17
M+25% Vetch in broadcast	0.20	0.98	1.18
M+50% Vetch in broadcast	0.27	1.02	1.29
M+75% Vetch in broadcast	0.28	0.91	1.19
M+25% Lablab in row	0.13	0.92	1.05
M+50% Lablab in row	0.19	0.94	1.13
M+75% Lablab in row	0.25	0.87	1.12
M+25% Lablab in broadcast	0.13	0.98	1.11
M+50% Lablab in broadcast	0.21	0.98	1.19
M+75% Lablab in broadcast	0.19	1.02	1.21
LSD (P=0.05)	0.09	0.18	0.21
F-test	**	*	*
CV (%)	19.7	11.5	10.3

LSD = Least significant difference; CV = Coefficient of Variations; LERm=Partial LER of maize; LERf = Partial LER of forage legumes and Total LER=LERm+LERf

The total LER in all cases was more than unity showing that intercropping of the forage legumes with maize is more advantageous than sole cropping of maize. The highest total LER 1.46 was recorded when vetch was row-sown at 50% seed rate of its sole followed by vetch broadcast at the same rate (1.29) and lablab broadcast at 75% seed rate of its sole (1.21)(Table 7). These values indicated that intercropping gave a 45%, 29%, and 21% yield advantages than planting sole crops. Therefore, the total LER indicated that intercropping of maize and forage legumes was productive and had a yield advantage over the sole maize cropping. Interestingly, the first and the second highest yield advantages were obtained due to high dry biomass for intercropped vetch with only 10% sacrifice and 2% extra advantage of maize dry biomass yield, respectively. The yield advantage could be due to a possible efficient utilization of growth resources by the intercropped crops or the intercropping advantages of weed reduction, nitrogen fixation and increased light use efficiency (Willey, 1985; Reddy, 2000). Land equivalent ratio greater than unity, has been reported in sorghum/lablab (Ibrahim *et al.*, 1993) and maize/faba bean (Tilahun, 2002) intercropping. In other intercropping studies Berhanu (2004) reported stover a 32% yield advantage in oats/vetch intercropping. The highest advantage of growing maize mixtures was that the early maturing component of the mixtures leaves space for intercropping so that a food or feed legume could be effectively intercropped without affecting the maize yield (Amede *et al.*, 2005). Higher LER in

intercropping than sole-cropping has also been reported in maize/soybean by Ullah *et al.* (2007). From these results, it can be concluded that additional forage can be produced by intercropping suitable forage legumes at their appropriate methods and rates of sowing with a little or no sacrifice in maize yield.

#### 4. Conclusion

In general, this study, demonstrated that integration of forage legumes (lablab and vetch) into maize as an intercropping system can increase productivity per unit of land, enable additional forage crop production without significant sacrifice of maize grain and stover yield and can improve the stover feed value. The grain yield reduction observed due to intercropping as a whole system was 9.5%, which is assumed to be acceptable by subsistence farmers. Vetch was found to be the more promising legume species to be integrated with maize than lablab. The highest maize grain yield, forage biomass, crude protein yield and partial and total LERs were obtained from maize/vetch intercropping in which vetch was sown in rows at 50% of its sole seed rate. Therefore, from this finding, it can be concluded that intercropping of vetch with maize in rows at 50% of its sole seed rate was superior and should be used by farmers in the study area to produce additional forage and improve the crude protein contents of stover for increasing productivity of livestock. However, there is a need to screen more types of potential forage legumes such as Desmodium species, alfalfa etc., for enhancing the yield and quality

of food and feed by intercropping with cereals and optimizing the methods and rates of their sowing.

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## Influence of Weed Dynamics on the Productivity of Common Bean (*Phaseolus vulgaris* L.) in Eastern Ethiopia

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**Abstract:** Common bean is an important cash crop for smallholder farmers in Ethiopia. However, its yield is constrained by weed infestations. Therefore, a study was conducted in 2012 cropping season at Haramaya and Hirna research fields eastern Ethiopia, to determine the critical periods of weed-crop competition and yield losses in common bean at the two sites. The experiment consisted of sixteen treatments in two sets, *i.e* one weed-free and one weedy set each comprising weed competition durations up to 10, 20, 30, 40, 50, 60 and 70 days after crop emergence and up to harvest). The experiment was laid out as a randomized complete block design with three replications for each set. The dominant weed species were *Galinsoga parviflora* and *Parthenium hysterophorus* with the highest relative densities of 26.7 and 39.8% at Haramaya and Hirna, respectively. With increasing duration of weed interference, weed dry weight, and the number of days of common bean plant required to reach physiological maturity were increased whereas the pods per plant, seeds per pod, hundred seed weight, grain yield, aboveground biomass, and harvest index of the common bean crop were reduced. At Haramaya and Hirna, uncontrolled weed growth significantly reduced common bean grain yield by 70 and 48%, respectively compared to the grain yield obtained from the weed-free check plots. In conclusion, the results of the study revealed that, to reduce the loss in the grain yield of common bean by more than 10%, it is important to keep the crop weed-free between 140 to 608 growing degree days (24 to 70 days after crop emergence) at Haramaya and from 140 to 707 growing degree days (14 to 70 days after crop emergence) at Hirna.

**Keywords:** Critical period; *Phaseolus vulgaris* L.; Weed-crop competition; Yield loss

### 1. Introduction

Common bean (*Phaseolus vulgaris* L.) is an important crop for Ethiopia and is produced on about 331,708.15 ha of land in the country; with a total production of 387,802.3 tons (t) with an average yield of 1.17 t ha<sup>-1</sup> (CSA, 2012). It is often grown as a cash crop by smallholder farmers and used as a major food legume in many parts of the country where it is consumed in different types of traditional dishes (Habtu, 1994).

Nevertheless, the yield of the crop is limited by many biotic and abiotic factors among which weeds are the major constraints. Uncontrolled weed populations can substantially reduce the yield of the crop up to 90% (Tilahun, 1998). The yield losses can be reduced successfully by maintaining the fields weed-free during the critical period of weed crop competition (CPWC) (Swanton and Weise, 1991).

Critical periods of weed-crop competition are an important principle of integrated weed management (IWM) program. It is a period in the crop growth cycle during which weeds must be controlled to prevent yield losses (Knezevic *et al.*, 2002). Therefore, weeds that are present before or emerge after this period do not cause significant yield losses. Studies on the critical period of weed control are important in making weed control recommendations because they indicate the optimum time for implementing and maintaining weed control

and reduce cost of weed control practices (Hall *et al.*, 1992; Van Acker *et al.*, 1993).

Rezene and Kedir (2008) recommended twice hand weeding applied during 15-45 days after crop emergence for Mexican 142 variety of common bean but one early weeding applied during 20-25 days after crop emergence for two other apparently more competitive varieties of common bean 'Ex-Rico' and 'Red Wolaita' for the Central Rift Valley areas of Ethiopia, and twice hand weeding applied during 15-30 days after crop emergence for two common bean varieties, namely, Roba-1(improved) and Jimma local in Jimma area, western Ethiopia.

The competitive relationship between crop and weeds is highly dependent on many factors including, the characteristics of the crop, the weeds, the environmental variables, and the cultural practices (Knezevic *et al.*, 2002). To give more accurate information for growers, CPWC should be determined specifically for a particular region by considering the weed composition and climatic conditions (Knezevic *et al.*, 2002; Heshmati, 2007; Wu *et al.*, 2008). Therefore, this study was under taken to determine the critical period of weed-crop competition and yield loss in common bean under the growing conditions of eastern Ethiopia.

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## 2. Materials and Methods

### 2.1. Description of the Study Sites

The experiment was conducted during the 2012 main cropping season at Haramaya (09° 26' N latitude and 42° 3' E longitude, and altitude of 2006 meters above sea level) and Hirna (09° 15' N latitude and 41° 6' E longitude, and altitude of 1870 meters above sea level), in eastern Ethiopia. The soil of the experimental site at Haramaya had organic matter content of 1.0%, total nitrogen content of 0.17%, available phosphorus content of 8.72 mg kg soil<sup>-1</sup>, pH of 8.13 and per cent sand, silt and clay content of 63, 20 and 17, respectively with sandy loam texture (Bethelhem, 2012).

The soil of Hirna had organic matter content of 1.4%, total nitrogen content of 0.22%, available

phosphorus content of 32 mg kg soil<sup>-1</sup>, pH of 6.79 (Bethelhem, 2012). As stated by the same author the texture of the soil is pre dominantly clay, with, percent sand, silt, and clay contents of 27, 28 and 45, respectively.

Haramaya receives an average annual rainfall of 786.8 mm, with a mean annual temperature of 16.4°C; and Hirna receives an average annual rainfall of 966.9 mm, with a mean annual temperature of 19.1°C. Total rainfall during the cropping season (July-October) was 474 and 548 mm at Haramaya and Hirna, respectively. The mean minimum and maximum temperatures during the cropping season were 12 and 24°C at Haramaya, respectively. The corresponding records for Hirna were 13 and 27°C, respectively (Figure 1).

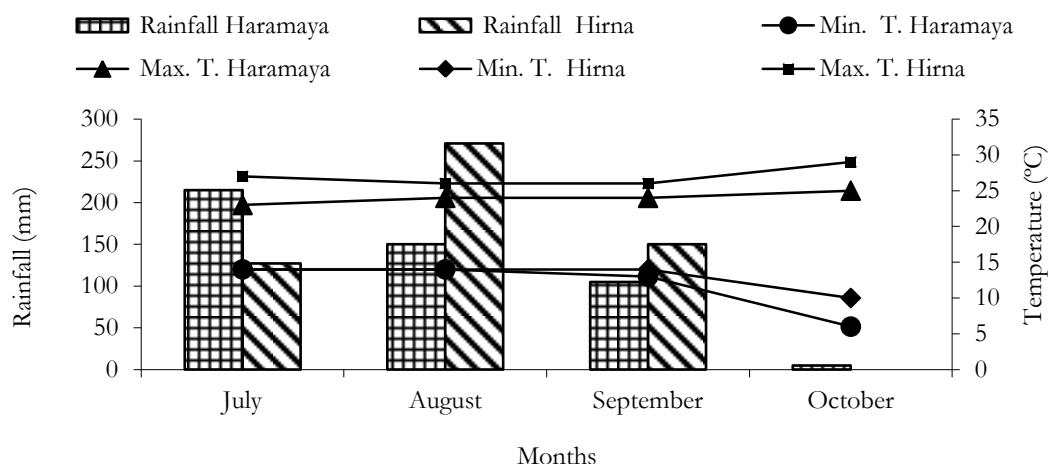


Figure 1. Rainfall, minimum and maximum temperatures recorded during the 2012 main cropping season at Haramaya and Hirna (Source: Jigijiga Meteorological Station).

### 2.2. Treatments and Experimental Design

The experiment consisted of sixteen treatments in two sets, *i.e.* one weed-free set and one weedy set each comprising weed competition durations up to 10, 20, 30, 40, 50, 60 and 70 days after crop emergence and up to harvest. The experiment was laid out as a randomized complete block design with three replications for each set. The experiment was arranged following the method described by Neito *et al.* (1968), and the treatments were compared with complete weed-free and weedy checks. The frequency of weeding the weed-free plots was based on the appearance of weeds. Growing degree days (GDD), which was used as an independent variable in regression analysis, was calculated as:

$$\text{GDD} = \sum (\text{Daily average temperature} - \text{Base temperature}).$$

The base temperature used in the calculation was 10°C (Hardwick, 1988).

### 2.3. Experimental procedure

The experimental field was prepared to seedbeds of a fine tilth. The gross plot size was 3.2 m x 2.4 m, with 8

rows spaced at the intra- and inter-row spacing of 10 and 40 cm, respectively, with a net harvestable area of 2.4 m x 1.6 m for each plot. Seed of the export type common bean variety Awash melka, which was released by Melkassa Agricultural Research Centre in 1998, was sown on 13 and 18 July 2012 at Hirna and Haramaya, respectively. Diammonium phosphate (18 kg N and 46 kg P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup>) was drilled in furrows at the recommended rate of 100 kg ha<sup>-1</sup> at planting. Weeds were removed by hoeing as required. Harvesting was done manually at harvest on 28 October 2012 and 6 November 2012 at Hirna and Haramaya, respectively. The biomass after harvest was sun-dried for 7-10 days and threshing and winnowing were done subsequently.

### 2.4. Data Collection

The weed flora present in the experimental fields were recorded from the weedy check plots in each replication just before flowering of the crop by placing a quadrat (0.25 m x 0.25 m) randomly at two spots in each replication, which was converted into m<sup>2</sup>. The species were categorized into their botanical families. Weed dry weight was determined from each plot during

weed removal for the early competition and about 15 days before final harvest for late competition to avoid possible foliage and seed shedding while throwing the quadrat (0.25 m × 0.25 m) randomly at two places. After three days of sun-drying, the samples were oven-dried at 65°C to a constant weight and their weights taken and subjected to square root transformation ( $\sqrt{x+0.5}$ ) to ensure normality before analysis.

Days to 90% physiological maturity was recorded in each plot, as the number of days from planting to when 90% of the 10 pre-tagged plant senesced and the leaves and pods turned yellow in color. The total number of pods in 10 randomly selected plants in each plot was counted at harvest and expressed as the number of pods per plant. From these pods, seeds were counted to determine the number of seeds per pod. Hundred seeds were counted from each plot, and their weight recorded. Aboveground dry biomass weight was measured at physiological maturity by cutting 10 randomly sampled plants at ground level and sun drying the biomass. This sun-dried aboveground biomass was multiplied by the number of plants in the net plot area to calculate the harvest index as the ratio of grain yield to the total aboveground dry biomass yield. Aboveground dry biomass was converted into ha.

Grain yield (kg) was recorded from each net plot area. The moisture content was determined for each treatment and adjusted at 10.5%. The maximum common bean yield loss due to weed competition was calculated as:

$$\left(1 - \frac{\text{Common bean yield in weedy check}}{\text{Common bean yield in weed-free check}}\right) \times 100 \quad (1)$$

## 2.5. Data Analysis

Data for each site were analyzed separately because of differences in weed composition, agro-climate, soil and planting date between the sites. The data were subjected to analysis of variance (ANOVA) and means were compared with the Least Significant Difference (LSD) test at 5% level of significance using SAS software program version 9.1 (SAS Institute, 2003).

To calculate the critical period of weed control in common bean, the relative common bean yield ( $Y$ ) of each treatment was calculated as:

$$\left(\frac{\text{Common bean yield in treatment}}{\text{Common bean yield in weed-free check}}\right) \times 100 \quad (2)$$

And non-linear regression equations were used to fit the data using STATISTICA software (StatSoft, 2004).

The onset and end of critical period, which is the duration mandatory for controlling weeds was estimated by the response curve when both curves attained 90% of the relative yield gain and 10% of the yield loss of the complete weed-free period. The critical period was determined and found to be in between these two threshold points.

Analysis was based on the models suggested by Knezevic *et al.* (2002). The Gompertz equation was used for describing the effect of increasing duration of weed control on common bean yield and the logistic equation was used for describing the effect of increasing duration of weedy period on common bean yield. The Gompertz equation used was:

$$Y = a \exp(-b \exp(-kT)) \quad (3)$$

Where  $Y$  is relative yield,  $a$  is the yield asymptote,  $b$  and  $k$  are constants, and  $T$  is the time (x-axis expressed in GDD). The logistic equation used was:

$$Y = \left[ \left( \frac{1}{\exp[c \times (T-d)] + f} \right) + \left[ \frac{(f-1)}{f} \right] \right] \times 100 \quad (4)$$

Where  $Y$  is relative yield,  $T$  is the time (x-axis expressed in GDD);  $d$  is the point of inflection,  $c$  and  $f$  are constants.

## 3. Results and Discussion

### 3.1. Weed Parameters

#### 3.1.1. Weed Flora in the Experimental Sites

Weed species diversity was more at Haramaya than Hirna (Table 1). The weed community was composed of 10 and 5 species at Haramaya and Hirna, respectively. Overall weed density ranged up to 101 and 94 weeds m<sup>-2</sup> at Haramaya and Hirna, respectively.

Table 1. Relative density (plants m<sup>-2</sup>) and percent (%) of weed species in the experimental fields of common bean at Haramaya and Hirna during 2012 main cropping season.

Weed species	Family	Haramaya		Hirna	
		Density (m <sup>-2</sup> )	(%)	Density (m <sup>-2</sup> )	(%)
<b>Broadleaved</b>					
<i>Amaranthus dubius</i> TheIl.	Amaranthaceae	-	-	16	6.8
<i>Argemone mexicana</i> L.	Papaveraceae	16	4.2	-	-
<i>Commelina benghalensis</i> L.	Commelinaceae	24	6.4	20	8.5
<i>Equisetum arvense</i> L.	Equisetaceae	29	7.7	-	-
<i>Galinsoga parviflora</i> Cav.	Asteraceae	101	26.77	32	13.5
<i>Medicago polymorpha</i> L.	Fabaceae	16	4.2	-	-
<i>Parthenium hysterophorus</i> L.	Asteraceae	-	-	94	39.8
<i>Plantago lanceolata</i> L.	Plantaginaceae	45	11.9	-	-
<i>Solanum nigrum</i> L.	Solanaceae	16	4.2	-	-
Total			65.3		68.6
<b>Grasses</b>					
<i>Dinebra retroflexa</i> (Vahl.) Panze	Poaceae	24	6.4	-	-
<i>Eragrostis cilianensis</i> (All.) Lut.	Poaceae	48	12.7	-	-
Total			19.1		-
<b>Sedge</b>					
<i>Cyperus rotundus</i> L.	Cyperaceae	59	15.6	74	31.4

The probable reason for more species occurrence at Haramaya could be the difference in soil type, previous crop (maize at Haramaya and wheat at Hirna), and more rainfall relative to Hirna at early stage of the crop growth (Figure 1). In line with this result, Tamado and Milberg (2000) reported that altitude, rainfall, month of planting, number of weeding and soil type were the major environmental/crop management factors that influenced weed species distribution.

*G. parviflora* and *C. rotundus* were the major weed species which were present at both sites. *P. hysterophorus* was the major weed species at Hirna. *G. parviflora* followed by *C. rotundus* were the dominant weed species at Haramaya whereas *P. hysterophorus* followed by *C. rotundus* were the dominant weed species at Hirna. The broadleaved, grass and sedge weeds constituted 65.3, 19.1 and 15.6% relative densities, at Haramaya and 68.6, 0 and 31.4% at Hirna, respectively (Table 1).

### 3.1.2. Weed Dry Weight

Significant differences were observed among the durations of weed competition on weed dry weight (Table 2). In general, weed dry weight increased with the increasing duration of the weedy period (IDWP) and decreased with the increasing duration of the weed-free period (IDWFP). In IDWP, the weeds might have exerted a severe competition and utilized the environmental resources for a longer period of time thus accumulating more dry matter. While in IDWFP, the weeds emerged and grew after the respective weed-free periods under stress, thus, accumulating lower dry weight. The lowest weed dry weight (gm<sup>-2</sup>) was found at 10 DAE under IDWP with the values of 12.3 and

18.4 gm<sup>-2</sup> at Haramaya and Hirna, respectively. But the value at Hirna was in statistical parity with the value obtained at 20 DAE (29.1 gm<sup>-2</sup>).

The highest weed dry weight found in the weedy check at Haramaya was in statistical parity with the values obtained 60 and 70 DAE while at Hirna a statistical parity occurred with the value obtained at 70 DAE (Table 2). Similarly, Ahmadi *et al.* (2007) reported that prolonging the weedy period in common bean increased the weed dry weight per unit area and caused an uninterrupted weed infestation throughout the crop growth period, resulting in the highest weed dry weight.

In IDWFP, the highest weed dry weight was found at 10 DAE with the values 290.4 and 545.3 gm<sup>-2</sup> at Haramaya and Hirna, respectively, which was drastically reduced as the weed-free period was prolonged at both sites (Table 2). The possible reasons for the higher weed dry weight at Hirna than at Haramaya could be the relatively higher rainfall and temperature at the latter during the cropping season, which may have induced more accumulation of weed dry matter (Figure 1; Table 2). Moreover, the soil of Hirna is more fertile than that of Haramaya, which may have favored weed growth. Corroborating the results of this study, Grundy and Mead (2000) also reported that high amounts of rainfall and temperature influenced the periodicity of weed emergence, which often resulted in increased weed dry weight. The differences in weed composition and abundance between the two sites could be attributed to differences in weather, soil and previous management practices used (Chikoye and Ekeleme, 2001; Knezevic *et al.*, 2002).

Table 2. Effect of increasing duration of weedy and weed-free periods on weed dry weight, days to physiological maturity, and number of pods per plant of common bean at Haramaya and Hirna during the 2012 main cropping season.

DAE	Weed dry weight (g m <sup>-2</sup> )		Days to physiological maturity		Number of pods per plant	
	Haramaya	Hirna	Haramaya	Hirna	Haramaya	Hirna
IDWP						
10	3.6 <sup>gh</sup> (12.3)	4.3 <sup>fg</sup> (18.4)	94 <sup>cd</sup>	89 <sup>cd</sup>	19.0 <sup>abc</sup>	24.8 <sup>ab</sup>
20	5.7 <sup>fg</sup> (31.7)	5.4 <sup>f</sup> (29.1)	94 <sup>cd</sup>	89 <sup>cd</sup>	21.9 <sup>abc</sup>	24.9 <sup>ab</sup>
30	7.5 <sup>ef</sup> (56.5)	10.7 <sup>de</sup> (114.1)	94 <sup>cd</sup>	90 <sup>bcd</sup>	18.4 <sup>abc</sup>	19.9 <sup>b-c</sup>
40	14.6 <sup>bc</sup> (212.5)	13.9 <sup>dc</sup> (194.1)	95 <sup>bcd</sup>	90 <sup>bcd</sup>	17.7 <sup>bcd</sup>	19.5 <sup>b-c</sup>
50	15.1 <sup>bc</sup> (231.0)	21.1 <sup>c</sup> (448.3)	95 <sup>bcd</sup>	90 <sup>bcd</sup>	12.7 <sup>de</sup>	19.2 <sup>b-c</sup>
60	21.5 <sup>a</sup> (465.1)	23.8 <sup>bc</sup> (571.5)	97 <sup>abc</sup>	92 <sup>abc</sup>	11.3 <sup>e</sup>	17.8 <sup>cde</sup>
70	21.8 <sup>a</sup> (475.7)	26.5 <sup>ab</sup> (700.5)	98 <sup>ab</sup>	93 <sup>ab</sup>	10.9 <sup>e</sup>	16.5 <sup>de</sup>
WC	23.0 <sup>a</sup> (528.5)	29.5 <sup>a</sup> (872.0)	99 <sup>a</sup>	94 <sup>a</sup>	9.9 <sup>e</sup>	15.8 <sup>e</sup>
IDWFP						
10	16.9 <sup>b</sup> (290.4)	23.2 <sup>bc</sup> (545.3)	99 <sup>a</sup>	94 <sup>a</sup>	17.0 <sup>cd</sup>	18.9 <sup>b-e</sup>
20	11.8 <sup>cd</sup> (153.1)	21.2 <sup>c</sup> (448.5)	98 <sup>ab</sup>	93 <sup>ab</sup>	18.1 <sup>bcd</sup>	20.1 <sup>b-e</sup>
30	11.6 <sup>cd</sup> (136.3)	14.7 <sup>d</sup> (217.1)	97 <sup>abc</sup>	92 <sup>abc</sup>	20.1 <sup>abc</sup>	21.2 <sup>a-e</sup>
40	9.8 <sup>ed</sup> (102.1)	9.9 <sup>e</sup> (145.1)	97 <sup>abc</sup>	92 <sup>abc</sup>	21.7 <sup>abc</sup>	22.3 <sup>a-d</sup>
50	8.3 <sup>def</sup> (73.9)	0.7 <sup>g</sup> (0.0)	97 <sup>abc</sup>	92 <sup>abc</sup>	22.3 <sup>abc</sup>	23.1 <sup>abc</sup>
60	5.7 <sup>fg</sup> (32.8)	0.7 <sup>g</sup> (0.0)	97 <sup>abc</sup>	92 <sup>abc</sup>	19.5 <sup>abc</sup>	22.0 <sup>a-d</sup>
70	0.7 <sup>h</sup> (0.0)	0.7 <sup>g</sup> (0.0)	97 <sup>abc</sup>	92 <sup>abc</sup>	22.9 <sup>ab</sup>	23.3 <sup>abc</sup>
WFC	0.7 <sup>h</sup> (0.0)	0.7 <sup>g</sup> (0.0)	93 <sup>d</sup>	89 <sup>cd</sup>	23.8 <sup>a</sup>	27.0 <sup>a</sup>
LSD <sub>(0.05)</sub>	3.6	4.4	3	3	5.5	6.2
CV(%)	19.2	20.5	1.8	2.0	18.3	17.6

DAE = days after crop emergence; IDWP = Increasing duration of weedy period; WC = Weedy check; IDWFP = Increasing duration of weed-free period; WFC = Weed-free check; Figures in parentheses are the original and those outside the parentheses are the square root transformed values; Means followed by the same letters within each column are not significantly different

Weed dry weight decreased significantly with the successive increases in the weed-free period up to 60 and 40 DAE, at Haramaya and Hirna, respectively. After those days, there was no weed emergence at both sites. Similar result was reported by Brian *et al.* (1993) who observed that weed dry weight decreased as the weed free period was prolonged in an experiment conducted to determine the critical period of weed control in common bean.

### 3.2. Common Bean Component

#### 3.2.1. Days to Physiological Maturity

Significant differences were observed among the durations of weed competition in days to physiological maturity (Table 2). At Haramaya, weedy check under IDWP took the maximum days (99) to attain physiological maturity; however, it was in statistical parity with the weedy period up to 60 and 70 DAE. On the other hand, no significant differences existed in days to physiological maturity when weeds were allowed to grow up to 10 to 60 DAE. There was also no significant difference among weedy plots from 40 to 70 DAE. Similar to Haramaya, at Hirna weedy check under IDWP took the maximum days (94) to attain physiological maturity; however, it was in statistical parity with the weedy period up to 60 and 70 DAE. The weedy plots from 10 to 60 DAE were also in statistical parity among each other. Weedy plots up to 70 DAE were in statistical parity with the rest of the IDWP treatments except with 10 and 20 DAE treatment (Table 2).

In IDWFP treatments, common bean plants reached physiological maturity at the same time at both sites except plants in plots which were kept weed-free throughout the growth period. The plants in this treatment reached physiological maturity significantly earlier (93 days) than the plants in other IDWFP treatments (Table 2).

The days required to reach physiological maturity of common bean at Haramaya were relatively longer than those required at Hirna. The probable reason could be relatively high rainfall and temperature observed at Hirna which might have favored the growth and development of common bean enhancing the days to reach physiological maturity.

In general, with increasing IDWP and decreasing IDWFP, the days required to reach physiological maturity increased. This means that the days required to attain physiological maturity increased as the duration of weed interference was prolonged (Table 2). The shading of crop plants by the weed canopy might have reduced sun light radiation thus prolonging the vegetative growth resulting in delayed days to physiological maturity. This in turn might have reduced vegetative growth and delayed the transition to the reproductive period and physiological maturity of common bean. Similarly, Mitiku *et al.* (2012) reported that with increase in the dry weight of parthenium, weed dry weight increased, and the days required by the common bean plants to reach physiological maturity were delayed.

### 3.2.2. Number of Pods per Plant

Significant differences were observed in the number of pods per plant due to increasing weedy period at Haramaya (Table 2). In IDWP treatments, no significant differences were found between 10 to 40 DAE at both sites. Similarly, no significant differences were observed between 40 and 50 DAE at Haramaya. The weedy check had the lowest number of pods per plant (9.9), which was in statistical parity with the number of pods per plant from plots kept weedy from 50 to 60 DAE. On the other hand, at Hirna, there were no significant differences between IDWP up to 50 DAE. Furthermore, keeping the plots weed-free from 30 DAE to 70 DAE resulted in a number of pods per plant that was statistical parity with the number of pods per plant from weedy check (15.8).

At Haramaya, in IDWFP though the lowest number of pods per plant (17.0) was recorded in plots which were kept weed-free up to 10 DAE, it was not statistically different from the plots which were weed-free up to 60 DAE. It was also observed that the plots kept weed-free from 30 to 70 DAE were as comparable as WFC (23.8) in number of pods per plant. At Hirna, all the treatments in IDWFP were statistically at par except 10 and 20 DAE with the values of 18.9 and 20.1, respectively, which had significantly lower number of pods per plant than WFC. Common bean plants at Hirna had more number of pods per plant than at Haramaya which might be the positive influence of relatively high amount of rainfall, temperature and fertile soil at Hirna (Figure 1).

In general, in most of the treatments, number of pods per plant was increased as weed interference decreased and the vice versa. This could be due to increased weed dry weight as weedy period increased and vice versa (Table 2). In line with this result, Ahmadi *et al.* (2007) reported that number of pods per plants significantly increased with increasing length of weed-free period and decreased with increasing length of weed infested period in common bean. Brain *et al.*

(1993) also reported that number of pods per plant was reduced in common bean with increasing durations of weed interference after planting. In contrast to this result, Mukhtar (2012) reported that duration of weed interference did not significantly affect number of pods per plant of irrigated common bean which could be due to more supply of water that might have increased the competitive ability of the crop.

### 3.2.3. Number of Seeds per Pod

Significant differences were observed in number of seeds per pod at Haramaya but at Hirna it was not significant (Table 3). At Haramaya, in IDWP treatments, the highest number of seeds per pod (6.9) was obtained when the crop was kept weedy up to 40 DAE which was statistically at par with weedy from 10 DAE to 30 DAE. It was also revealed that keeping the plots weedy beyond 40 DAE decreased the number of seeds per pod which was statistically at par with WC (5.7) (Table 3). In agreement with this result, Mukhtar (2012) reported that number of seeds per pod increased as weed interference period decreased and weed-free period increased.

### 3.2.4. Hundred Seed Weight

Increasing duration of weed-free period treatments had significant effect on hundred seed weight at Haramaya, but not at Hirna among the treatments (Table 3). However, IDWP had no significant effect on hundred seed weight at Haramaya. In IDWFP, WFC had the highest hundred seed weight (17.12 g) at Haramaya. However, it did not differ significantly with weed-free up to 70 DAE.

Similar to the current result, Mukhtar (2012) also reported that hundred seed weight was decreased as weed interference increased in common bean. On the other hand, Burnside *et al.* (1998) stated that weed removal treatments had little effect on hundred seed weight of harvested common bean.

Table 3. Effect of increasing duration of weedy and weed-free periods on number of seeds per pod, hundred seed weight and grain yield of common bean at Haramaya and Hirna during 2012 main cropping season.

DAE	Number of seeds per pod		Hundred seed weight (g)		Grain yield (kg ha <sup>-1</sup> )	
	Haramaya	Hirna	Haramaya	Hirna	Haramaya	Hirna
<b>IDWP</b>						
10	6.7 <sup>abc</sup>	6.2	15.35 <sup>c</sup>	18.42	2438.2 <sup>a</sup>	3619.5 <sup>abc</sup>
20	6.6 <sup>abc</sup>	5.4	15.34 <sup>c</sup>	18.39	2341.7 <sup>abc</sup>	3476.1 <sup>abc</sup>
30	6.7 <sup>ab</sup>	5.7	15.73 <sup>bc</sup>	18.02	2260.5 <sup>abc</sup>	3392.7 <sup>abc</sup>
40	6.9 <sup>a</sup>	5.6	15.84 <sup>bc</sup>	18.21	2004.6 <sup>bcd</sup>	3336.2 <sup>a-d</sup>
50	5.9 <sup>d</sup>	5.4	15.64 <sup>bc</sup>	18.07	1558.6 <sup>ef</sup>	2949.2 <sup>c-f</sup>
60	6.0 <sup>cd</sup>	5.3	15.78 <sup>bc</sup>	17.87	1325.8 <sup>fg</sup>	2500.6 <sup>def</sup>
70	6.1 <sup>bcd</sup>	5.6	15.23 <sup>c</sup>	17.83	1138.5 <sup>gh</sup>	2387.3 <sup>ef</sup>
WC	5.7 <sup>d</sup>	4.9	15.24 <sup>c</sup>	17.53	751.8 <sup>h</sup>	2174.8 <sup>f</sup>
<b>IDWFP</b>						
10	6.7 <sup>ab</sup>	5.7	15.93 <sup>bc</sup>	17.72	1575.7 <sup>ef</sup>	3128.4 <sup>b-e</sup>
20	6.4 <sup>a-d</sup>	5.5	14.89 <sup>c</sup>	17.85	1835.4 <sup>de</sup>	3333.3 <sup>a-d</sup>
30	6.9 <sup>a</sup>	5.5	14.95 <sup>c</sup>	17.87	1970.6 <sup>cde</sup>	3387.2 <sup>abc</sup>
40	6.87 <sup>a</sup>	5.6	15.30 <sup>c</sup>	18.05	2005.0 <sup>bcd</sup>	3488.8 <sup>abc</sup>
50	6.6 <sup>abc</sup>	6.1	14.93 <sup>c</sup>	18.34	2084.4 <sup>a-d</sup>	3529.4 <sup>abc</sup>
60	6.7 <sup>abc</sup>	6.0	15.14 <sup>c</sup>	17.92	2265.1 <sup>abc</sup>	3819.3 <sup>ab</sup>
70	6.7 <sup>abc</sup>	5.9	16.58 <sup>ab</sup>	18.86	2416.7 <sup>ab</sup>	3925.3 <sup>ab</sup>
WFC	6.7 <sup>ab</sup>	6.0	17.12 <sup>a</sup>	18.89	2468.1 <sup>a</sup>	4181.3 <sup>a</sup>
LSD <sub>(0.05)</sub>	0.7	NS	1.06	NS	413.7	849.7
CV(%)	6.3	7.6	4.1	3.7	13.0	15.5

DAE = days after crop emergence; IDWP = Increasing duration of weedy period; WC = Weedy check; IDWFP = Increasing duration of weed-free period; WFC = Weed-free check; NS = not significant; Means followed by the same letters within each column are not significantly different

### 3.2.5. Grain Yield

Common bean grain yield varied significantly with the variations in the duration of competition. At both sites, the grain yield decreased with the increase in the duration of weedy periods and with the decrease in the duration of weed-free periods (Table 3). However, the extent of reduction was remarkable beyond weedy periods of 30 and 40 DAE at Haramaya and Hirna, respectively.

At Haramaya, there were no significant differences in the grain yield with weed free check (WFC) when the weedy period extended from 10 to 30 DAE and the weed free periods panned from 50 to 70 DAE. A similar trend was observed between the weedy periods from 20 to 40 DAE while beyond this period the yield declined significantly. In contrast, at Hirna, no significant yield difference was obtained when the weedy period was extended up to 40 DAE, and beyond this period, the grain yield was significantly reduced.

The yield obtained in weed-free check plots at Hirna was in statistical parity with the yield obtained during the time spanning from 10 to 40 DAE and 20 to 70 DAE, respectively, under increasing duration of weedy period and increasing duration of weed-free period treatments. The weedy check plots also produced the lowest grain yield at Hirna, but the yield did not differ significantly with the yield obtained from the plots that remained weedy from 50 to 70 DAE. The decrease in yield with the increase in the duration of competition

might be the result of increased weed dry weight, which might have influenced the number of pods per plant. Inconformity with this result, Burnside *et al.* (1998), Ahmadi *et al.* (2007), and Mukhtar (2012) reported the important common bean weed competition periods to be between 3 to 6 weeks after sowing, 19 to 52 DAE and 4<sup>th</sup> to 6<sup>th</sup> weeks after sowing, respectively.

Decreasing the duration of weed competition revealed no significant differences in grain yield between weed-free check plots and weed free plots up to 50 to 70 DAE at Haramaya. However, at Hirna weed-free check plots had significant difference with plots kept weed free up to 10 DAE. Comparing the two sites, the highest common bean yield was recorded at Hirna whereas the lowest yield was obtained from Haramaya (Table 3). This might be attributed to the better soil fertility status, relatively higher rainfall, and warmer temperature that is more conducive for growth and development of the crop at the former than the latter (Figure 1).

The common bean yield losses in the weedy checks as compared to the weed-free checks were 70 and 48% at Haramaya and Hirna, respectively (Table 4). The higher yield loss at Haramaya could be ascribed to more number of weed species (Table 1), relatively low seasonal rainfall and temperature (Figure 1) and relatively lower soil fertility compared to Hirna. Consistent with the results of this study, common bean yield losses of 98, 69.9, and 57.98% were reported by

Ahmadi *et al.* (2007); Dawit *et al.* (2011) and Mukhtar (2012), respectively, in weedy check plots as compared to the weed-free check plots.

### 3.2.6. Aboveground Dry Biomass Yield

The aboveground dry biomass yield of common bean was significantly influenced both by the increasing and decreasing periods of weed competition at Haramaya but it was not significantly influenced at Hirna (Table 4). At Haramaya, in IDWP treatments, no significant difference was found between 10 to 40 DAE. Similarly, no significant difference was observed between 50 to 70 DAE at Haramaya. The aboveground dry biomass

yield of the weedy check plot, which was the lowest, was in statistical parity with the aboveground dry biomass yield obtained from plots which were kept weedy for 50 and 70 DAE.

At Haramaya, in IDWFP the lowest dry biomass (3819.4 kg ha<sup>-1</sup>) was recorded in plots which were kept weed-free up to 10 DAE. This value was in statistical parity with the plots which were weed-free up to 60 DAE except with 40 DAE treatments. It was also observed that, the plots that were kept weed free from 60 to 70 DAE were not significantly different from WFC in the amount of aboveground dry biomass produced.

Table 4. Effect of increasing duration of weedy and weed-free periods on aboveground dry biomass, harvest index and yield loss of common bean at Haramaya and Hirna during 2012 main cropping season.

DAE	Aboveground dry biomass(kgha <sup>-1</sup> )		Harvest index (%)		Yield loss(%)	
	Haramaya	Hirna	Haramaya	Hirna	Haramayaaaya	Hirna
IDWP						
10	5208.3 <sup>ab</sup>	7476.4	46.8 <sup>a</sup>	48.4	1	13
20	5902.8 <sup>a</sup>	7302.8	39.7 <sup>abc</sup>	47.7	5	17
30	5902.8 <sup>a</sup>	6955.6	38.3 <sup>abc</sup>	48.7	8	19
40	5208.3 <sup>ab</sup>	6781.9	38.6 <sup>abc</sup>	49.2	19	20
50	3993.1 <sup>cd</sup>	6261.1	39.0 <sup>abc</sup>	47.9	37	29
60	4340.3 <sup>bc</sup>	6000.7	30.5 <sup>bcd</sup>	41.6	46	40
70	3819.4 <sup>cd</sup>	5827.1	29.6 <sup>cd</sup>	42.2	54	43
WC	3125.0 <sup>d</sup>	5566.7	24.1 <sup>d</sup>	39.9	70	48
IDWFP						
10	3819.4 <sup>cd</sup>	6087.5	41.6 <sup>abc</sup>	52.2	36	25
20	4513.9 <sup>bc</sup>	7042.4	40.7 <sup>abc</sup>	47.7	26	20
30	4687.5 <sup>bc</sup>	6608.3	42.4 <sup>ab</sup>	51.3	20	19
40	5208.3 <sup>ab</sup>	6521.5	38.5 <sup>abc</sup>	54.5	19	17
50	4687.5 <sup>bc</sup>	6608.3	44.5 <sup>a</sup>	54.4	16	16
60	4861.1 <sup>abc</sup>	6955.6	46.8 <sup>a</sup>	54.9	8	9
70	5902.8 <sup>a</sup>	7389.6	41.0 <sup>abc</sup>	54.1	2	6
WFC	5902.8 <sup>a</sup>	7476.4	42.2 <sup>ab</sup>	55.9	0	0
LSD <sub>(0.05)</sub>	1090.4	NS	12.2	NS	-	-
CV(%)	13.6	14.9	18.6	21.3	-	-

DAE = days after emergence; IDWP = Increasing duration of weedy period; WC = Weedy check; IDWFP = Increasing duration of weed-free period; WFC = Weed-free check; NS = not significant; Means followed by the same letters within each column are not significantly different

In general, at Haramaya, the aboveground dry biomass of common bean was decreased with the increasing duration of weedy periods and with decreasing durations of weed-free periods (Table 4). However, the rates of reductions were remarkable beyond weedy periods of 40 DAE at Haramaya. Furthermore, the rate of reduction in weed-free period was remarkable before weed-free period of 50 DAE except in 40 DAE. The highest biomass yield (5902.8 kg ha<sup>-1</sup>) was obtained from weed-free check plots at Haramaya.

### 3.2.7. Harvest Index

Harvest index was significantly affected by the duration of weed competition at Haramaya but not at Hirna (Table 4). At Haramaya, in IDWP treatments, no

significant difference was found between 10 to 50 DAE. Similarly, no significant difference was observed between 50 and 60 DAE. The mean harvest index of the weedy check plots, which was the lowest (24.1%), was in statistical parity with the harvest index obtained in plots kept weedy for 60 and 70 DAE. In IDWFP treatments, harvest index was not significantly affected by the treatment. The common bean harvest index at Hirna exceeded the one at Haramaya. This may be ascribed to the better edaphic and weather conditions for growth and development of the crop at Hirna than Haramaya (Figure 1). The favorable conditions at Hirna might have helped the common bean plants to produce and partition more total dry matter yield into the economic yield.

### 3.2.8. Critical Periods of Weed Control

The Gompertz and logistic equations generally described the data well as indicated by high coefficients of determination ( $R^2$ ) values (Table 5). The predicted and the observed relative common bean yield as affected by duration of the weed-free and weed infested periods are shown in Figure 2.

The results showed that in order to avoid more than 10% common bean grain yield loss, the maximum time that weeds could be allowed to infest after planting the crop (the beginning of the critical period) were about 240 GDD (24 DAE) and 140 GDD (14 DAE) at Haramaya and Hirna, respectively (Figure 2).

Table 5. Parameter estimates for the Gompertz and logistic equations.

Sites	Gompertz parameters				Logistic parameters			
	A	b	k	$R^2$	c	d	f	$R^2$
Haramaya	89.94	0.99	0.0096	0.97	0.05	0.0055	1.07	0.99
Hirna	89.89	0.50	0.0070	0.95	0.37	0.0018	1.32	0.97

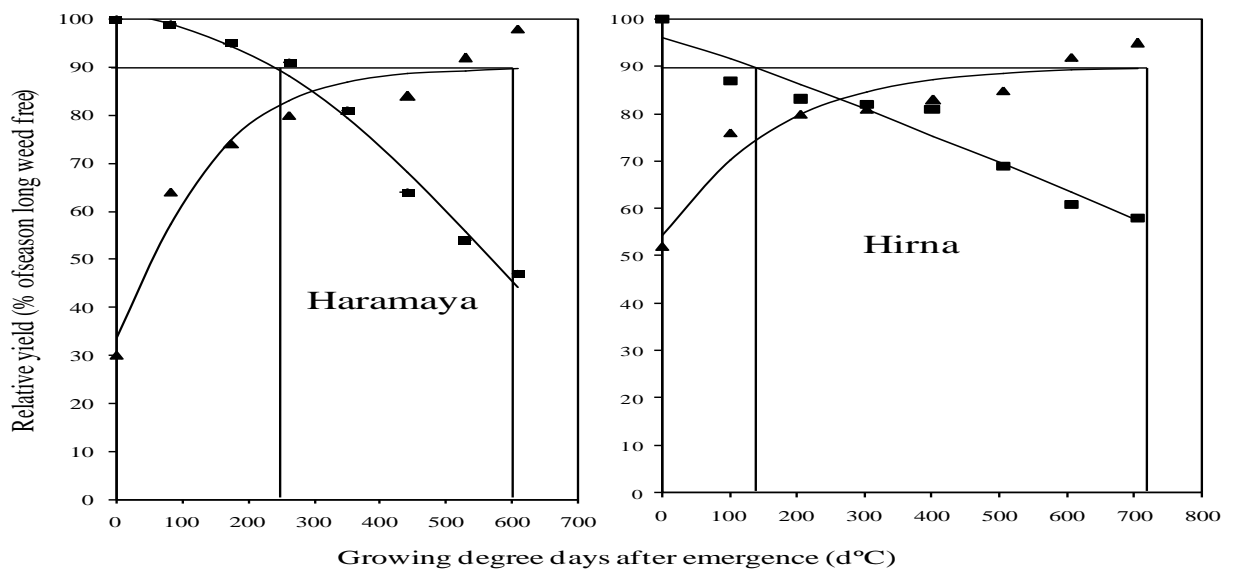


Figure 2. Effect of weed interference on relative common bean yield at Haramaya and Hirna during 2012 main cropping season.

Increasing the duration of weed interference (■) and fitted curve as calculated by the logistic equation; increasing weed-free period (▲) and fitted curve as calculated by the Gompertz equation. Horizontal line indicate the 10% acceptable yield loss; vertical lines indicate the starting and end of CPWC.

The earlier start of the critical period of weed interference at Hirna could be attributed to the higher minimum and maximum temperatures (Figure 1) that might have resulted in early emergence, establishment and rapid growth of weeds thus utilizing the available resources more efficiently and posing a stiffer competition of the weeds with the crop for growth resources (Table 2). Consistent with this suggestion, Gupta (2011) reported that the weeds that germinated earlier, before or at the same time as the crop emergence, posed a serious competition to the crop plants since they had an opportunity to establish and accumulate dry matter faster than the crop plants.

Knezevic *et al.* (2002) also reported that the critical period of weed interference for a given crop can vary with the relative time of weed emergence, because earlier weed emergence can lead to the earlier beginning of the critical period. In conformity with this result, Hall *et al.* (1992) also reported that weed density appears to be important in the determination of the beginning of the critical period where the critical period tended to start later for experiments with lower weed density in maize. At very low weed densities there might be even no critical period of weed interference (Van Acker *et al.*, 1993; Martin *et al.*, 2001). Likewise, Lindquist *et al.* (1999) indicated that the relative time of weed and crop emergence and densities of both crop and weed might explain the variation in crop-weed interference relationship among sites.

In this study, the end of the critical period of weed interference at the 10% acceptable common bean grain yield loss level were about 608 GDD (70 DAE) and

707 GDD (70 DAE) at Haramaya and Hirna, respectively. It lasted almost until the end of the crop growing season at both sites. Long lasting CPWC at Hirna might be due to the dominance of more competitive weeds such as *P. hysterophorus*, *C. rotundus* and *G. parviflora* (Table 1). In line with this result, Arslan *et al.* (2006) reported that long lasting CPWC might result in more competitive weeds such as Johnson grass (*Sorghum halepense* L.) and common cocklebur (*Xanthium strumarium* L.) during determination of critical period of double cropped soybean.

Critical period of weed crop competition obtained in this study was narrower at Haramaya while it was wider at Hirna as compared to the result reported by Ahmadi *et al.* (2007), who found that the critical period of common bean ranged from 206 to 745 GDD (approximately 19 to 52 DAE) at an acceptable yield loss of 10%. This variation could be explained by differences in environmental conditions, management practices, weed species diversity *etc.* as the CPWC has been found to vary with location, year, weed species, relative time of weed emergence, weed density, cultivar, agronomic practices *etc.* (Van Acker *et al.*, 1993; Knezevic *et al.*, 2002).

#### 4. Conclusion

The results of this study indicated that the maximum common bean yield losses due to the highest weed interference were 70% and 48% at Haramaya and Hirna, respectively, as compared to the weed free check. To prevent more than 10% yield loss, the efficient weed control methods for common bean variety Awash Melka could be accomplished by keeping the crop weed free between 140 to 608 GDD (24 to 70 DAE) at Haramaya and 140 to 707 GDD 14 to 70 DAE at Hirna. This could be done by using cultural, chemical, and integrated weed management practices.

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## Farmer Participatory Evaluation of Agronomic Performances of Bread Wheat Varieties in the Highlands of Eastern Ethiopia

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**Abstract:** Although a number of improved wheat varieties have been released in Ethiopia, most farmers continue cultivating local varieties, which are low yielders and highly susceptible to diseases. The low adoption rate of improved wheat varieties is attributable mainly to farmers' uncertainty about the expected benefit. Hence, on-farm trials consisting of three improved bread wheat varieties [Madda Walabu (HAR-1480), Digalu (HAR-3116), Danda'a], with one local variety as a control treatment, were conducted in Gurawa district in the highlands of eastern Ethiopia in the 2012 and 2013 cropping seasons. The objective of the trials was to evaluate the varieties for agronomic performances jointly with farmers' research groups, researchers, and experts of agriculture in the region. Visual observation and data collection were done from planting up to harvesting. The visual assessment revealed that the growth performances of the improved varieties were superior to that of the local variety. The analysis of variance showed that the grain yields of the improved varieties significantly exceeded that of the local variety. Thus, the improved varieties Digalu, Danda'a, and Madda Walabu produced grain yields of 7432.5, 7193.7, 6502.5 kg ha<sup>-1</sup>, which exceeded the grain yield produced by the local variety (4835 kg ha<sup>-1</sup>) by about 54, 49, and 34%, respectively. Digalu and Danda'a were also unaffected by diseases compared to the other varieties. Eventually, the farmers selected Digalu and Danda'a, but rejected the improved Madda Walabu variety and the local one. It could be concluded that farmer participatory evaluation of existing improved wheat varieties is a vital pre-condition for adoption and scaling up of production of the crop in the region.

**Keywords:** Disease resistance; Improved varieties; Local variety; *Triticum aestivum* L.; Yield

### 1. Introduction

Wheat is one of the most important cereal crops of the world and is a staple food for about one third of the world's population (Hussain and Shah, 2002). It is a major cereal crop in Ethiopia, which is largely grown in the highlands. At the national level, wheat is cultivated on 1.63 million ha of land with a total grain production of 3.43 million tonnes (CSA, 2013), and the country is considered the largest producer of the crop in sub-Saharan Africa. Bread wheat (*Triticum aestivum* L.) accounts for about 60% of the total wheat production in the country whereas durum wheat (*Triticum aestivum* L.) accounts for the remaining production (Hailu, 1991). Bread wheat is preferred to durum wheat by farmers in Ethiopia owing to its high yield potential, ease of mechanization, relatively higher economic returns, and good bread making quality relative to the other food crops (Tanner *et al.*, 1993).

However, one challenge faced in wheat production in the country is low productivity per unit area of land. The national average yield of the crop is estimated at 2.11 tonnes ha<sup>-1</sup> (CSA, 2013), which is very low compared to the world's average yield of 3.09 tonnes ha<sup>-1</sup> (FAOSTAT, 2012). Low productivity of the crop is attributed to the use of old and low-yielding varieties, depletion of soil nutrients, poor weed management practices, low levels of fertilizer application,

waterlogging in vertisol areas, prevalence of aggressive and virulent crop pathogens, and unavailability of modern crop management inputs (Tanner *et al.*, 1993). Moreover, the conventional research in the country hardly involves farmers in a participatory approach in both problem diagnosis and evaluation of research results. Consequently, improved technologies have suffered lower rates of adoption. This is because farmers' real problems and their selection criteria are not considered in the research process (Witcombe *et al.*, 1996; Witcombe *et al.*, 2005; Abebe *et al.*, 2013).

Participatory Rural Appraisal (PRA) and scoping studies were conducted in eastern Ethiopia by the CASCAPE (Capacity Building for Scaling up of Evidence-Based Best Practices in Agricultural Production in Ethiopia) project, which is being implemented by Haramaya University in collaboration with Alterra-Wageningen UR. During the implementation of the project, cultivation of low yielding and disease-susceptible varieties and poor agronomic practices were identified as the major constraints to wheat production in the highlands of eastern Ethiopia (Nigussie *et al.* 2012). As a result, despite the suitability of the agro-ecological conditions, wheat production in the region has remained low.

Although a number of high-yielding wheat varieties have been released through the research system in the country, farmers in Ethiopia are reluctant to adopt them, with only 3-5% of the cultivated land covered

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with seeds of improved crop varieties (World Bank, 2005). Shortage of seed of improved varieties, lack of attributes desired by farmers, low information exchange between farmers and researchers about the improved varieties, and farmers' uncertainty about the improved varieties or risk-aversion could be the major reasons for the low adoption rate of improved crop varieties (Wale and Yallew, 2007; Abebe *et al.*, 2013).

Increasing the adoption rate of improved crop varieties to enhance food production requires on-farm participatory breeding, evaluation, and demonstration of improved and high yielding crop varieties with recommended technological packages (Witcombe *et al.*, 1996; Witcombe *et al.*, 2005).

No participatory field evaluation of bread wheat varieties has been undertaken in the eastern highlands of Ethiopia to elucidate the problem of low adoption rate of the improved varieties. The objective of this research was, therefore, to evaluate the agronomic performances of existing improved bread wheat varieties by involving farmers in the process.

## 2. Materials and Methods

### 2.1. Experimental Site

Field trials were conducted in Gurawa district of the East Hararghe Zone of the Oromia Regional State during the 2012 and 2013 cropping seasons. Gurawa is one of the districts located in the highlands with high potential for wheat production in eastern Ethiopia. The altitude of the study sites is 2355 metres above sea level. The geographical location is 09°10'51.7"N latitude and 41°47'29.3"E longitude. The region is generally characterised by a mixed crop-livestock farming system. However, the animal production component is limited to the holding of small numbers of heads of cattle, sheep, and other livestock. Farmers in the study area produce mostly staple crops, namely, wheat, barley, potato, maize and faba bean.

### 2.2. Experimental Materials

Three improved bread wheat varieties [Madda Walabu (HAR-1480), Digalu (HAR-3116), Danda'a], and one local bread wheat variety were used for the experiment. The varieties are semi-dwarf. Madda Walabu was released by Sinana Agricultural Research Centre (SARC) in 2000 while Digalu and Danda'a were released by Kulumsa Agricultural Research Centre in 2005 and 2010, respectively. The improved varieties were chosen because they are high yielders and widely grown in the wheat belt highlands in the south-eastern part of the country. Early generation seeds of the improved varieties were obtained from SARC whereas that of the local variety was obtained from farmers. Nitrogen at the rate of 64 kg N ha<sup>-1</sup> in the form of urea (46% N) and phosphorus at the rate of 46 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> in the form of Di-ammonium phosphate (DAP) (20% P) were applied to all plots uniformly (Asnakew *et al.*, 1991).

### 2.3. Treatments and Experimental Design

The treatments consisted of the aforementioned four bread wheat varieties: Madda Walabu (HAR-1480), Digalu (HAR-3116), Danda'a and one local check. The experiment was conducted at four sites on four farmers' fields by involving groups of farmers. It was replicated four times following the procedure of randomized complete block using farm fields as blocks. The size of each plot was 10 m x 10 m and the distance between plots was 1.5 m.

### 2.4. Experimental Procedure

#### 2.4.1. Fertilizer application and field management

Land preparation was carried out following the standard production practices. Planting was done by uniformly drilling the seed into rows of 20 cm spacing at the recommended seed rate of 150 kg ha<sup>-1</sup>. Phosphate fertilizer was supplied equally to all plots as a basal application at the time of planting and covered with the soil. Nitrogen fertilizer was applied in two equal splits at planting and at Zadoks growth stage 23 (Zadoks *et al.*, 1974). For supplying N at planting, urea was applied as a side-dress and incorporated into the soil. The second split N fertilizer application as dry urea was done by top-dressing at the specified Zadoks growth stage. All broad leaf and grass weeds were removed by hand weeding two times.

#### 2.4.2. Site selection and organization of farmers' research groups

Site and farmer selections were carried out together with development agents (DAs) and woreda (district) experts. Before implementation of the activities, the selected groups of farmers were briefed on the process of participatory technology demonstration and evaluation as well as group learning and experience sharing. Consequently, the researchers together with the development workers identified and selected farmers and fields for the trials.

Four Farmers' Research Groups (FRGs), each consisting of 12-15 members, were organized for the trials at the planning time and given the task of carrying out field activities, monitoring, and evaluation. The FRGs were organized based on neighbourhood affinity and willingness to allot their time and land for implementation of the planned activities. Each group had one leader to facilitate the evaluation process with the other members. Equal opportunities were given to the community members to join the FRGs. In the group formation, the balance of age, gender, and wealth were kept. To enhance women's participation in the technology evaluation, 2-3 female-headed households were included in each group. Moreover, on-spot training on the production technologies was provided for the FRGs.

#### 2.4.3. Evaluation process by the FRGS and other stakeholders

Evaluation by the FRGs was undertaken at the critical growth stages of the crop and continued until the time

of harvesting. Farmers' rankings and preferences were observed over time for the varieties. There were pre-set time schedules prepared for evaluation and meeting sessions of the FRGs. Accordingly, evaluations of the trials were conducted at tillering (Zadoks 23) and maturity (Zadoks 93) stages. Farmers' evaluation and discussions were facilitated, and their criteria and preferences recorded by the DAs (Development agents) with close supervisions by researchers from the project. At tillering stage, farmers based their evaluation criteria on tillering capacity, stand vigour, leaf greenness, and disease resistance. Final evaluation was based on disease resistance and grain yields. In addition, seed susceptibility to sprouting, seed plumpness, and seed weight were used as evaluation criteria by the farmers at harvest.

Moreover, field days were organized for demonstration and evaluation purposes at the grain filling stage of the crop. During the events, experiences and knowledge were shared by farmers and other stakeholders (zonal and woreda offices of agriculture, regional agricultural research institute, university and NGOs) who are involved in agriculture and rural development activities in the region. Hence, differences in varietal performances in terms of growth, disease reaction, and yield were evaluated by a large group of participants.

## 2.5. Data Collection and Analysis

Plant growth and yield data were collected by the researchers. The data collected included days to physiological maturity, plant height, number of fertile spikes per m<sup>2</sup>, spike length, number of fertile tillers per plant, number of spikelet per spike, number of seeds per spikelet, number of seeds per spike, biomass yield (kg ha<sup>-1</sup>), grain yield (kg ha<sup>-1</sup>), and harvest index.

The number of days required by the wheat plants to reach physiological maturity was determined as the number of days required from sowing to the date when 50% of the peduncles senesced (turned yellow), i.e., when no green colour remained on the glumes and peduncles of the plants. Plant height (cm) was recorded as the average height of ten plants randomly sampled at physiological maturity. It was measured from the surface of the soil to the top of the spike excluding the awns. The number of fertile spikes per m<sup>2</sup> was determined at maturity by counting all fertile or seed-producing spikes from a 3 m<sup>2</sup> area of the central rows of each plot. The number of fertile tillers per plant was recorded as the average count of productive tillers of ten randomly selected plants at maturity.

Spike length (cm) was recorded as the average length of ten randomly sampled spikes, and measured from the base of the spike up to the apex of the terminal spikelet, excluding the awns. The number of spikelet spike<sup>-1</sup> was taken as the average number of separate spikelet of an individual rachis from ten randomly sampled spikes of the main culms. The number of seeds per spike was determined as the average number of kernels counted for ten spikes of the main culms.

The number of seeds per spikelet was determined as the quotient of the number of seeds per spike and number of spikelet per spike.

Biomass yield (kg ha<sup>-1</sup>) was measured at maturity by cutting all the plants in the central 3 m<sup>2</sup> area in each pot at the ground level with a sharp sickle and weighing after air drying. Grain yield was measured from the central 3 m<sup>2</sup> area of the plots. Following harvesting and threshing, the sample yields were weighed using a digital balance. Harvest Index was calculated as the ratio of grain yield to the total aboveground biomass yield. In addition to the biological data, farmers' perceptions and preferences were collected and narrated. Analysis of variance for the quantitative data was conducted using SAS GLM procedure (SAS Inst., 2004). Significant differences among the varieties were computed using the LSD test at 5% level of significance.

## 3. Results and Discussion

### 3.1. Evaluation by the Farmers' Research Groups (FRGs)

According to the farmers' evaluation, the three improved varieties (Madda Walabu, Danda'a and Digalu) performed best in terms of growth and yield compared to the local variety. Farmers' preferences for the improved varieties, however, varied due to varietal differences in terms of stand vigour, disease (rust) resistance and yield potential. At the seedling stage, variety Madda Walabu exhibited better stand vigour and rooting characteristics (dense and deep rooting). At the tillering stage, however, the FRGs preferred and ranked variety Digalu at the top of all other four varieties because of its stand uniformity and vigour, higher tillering capacity, leaf greenness, and resistance to the yellow rust disease (*Puccinia striiformis* f.sp. *tritici*) (Table 1). Similar studies conducted in Arsi Zone of Oromia and Tigray regions showed that varieties Digalu and Danda'a were preferred by farmers due to their higher resistance to yellow rust, stem rust, and *Septoria*, which occur persistently in the areas (EAAPP, 2012).

At maturity, the FRGs evaluation criteria focussed on varietal resistance to stem rust (*Puccinia graminis* f. sp. *tritici*), susceptibility to sprouting, number of seeds per spike, straw yield, grain yield, seed plumpness, and seed weight. According to the ranking set by the FRGs, variety Digalu stood first followed by Danda'a and Madda Walabu whilst the local variety was ranked the least. Despite its long spikes, variety Madda Walabu was outperformed by both Danda'a and Digalu in terms of number of seeds per spike. Moreover, this variety was found to be infected by stem rust while no disease symptoms were observed on the other two varieties, i.e., Digalu and Danda'a. Consistent with the results of this study, an assessment of wheat rust diseases recently conducted in eastern Ethiopia has revealed that Madda Walabu has in fact become moderately susceptible to yellow and stem rusts (FARC, 2013).

The farmers selected Digalu as the first, variety Danda'a as the second and variety Madda Walabu as the third best varieties in agronomic performances. The farmers did not select the local variety. In this study, most farmers preferred the late maturing variety Digalu whose maturity time coincided with the time of cessation of the rainy season. Late maturity imparts the variety with freedom from the likelihood of seed sprouting as a result of wet weather since the time of harvesting would coincide with the onset of the dry season. In addition, late maturing enhances the potential of the crop for increased grain filling, which results in higher yields (Al-Karaki, 2012). Similarly, in southern Ethiopia, the same variety (Digalu) was preferred by farmers mainly due to its potential for

high yield, disease resistance, and frost tolerance as well as high stand vigour and tillering capacity (Mathewos *et al.*, 2012).

Moreover, evaluation of the varieties at the grain filling stage by farmers during the field days helped to strengthen linkages and created opportunities for knowledge transfer among stakeholders. The occasion also provided the opportunity for different stakeholders to identify research and intervention gaps. The overall farmers' evaluation conducted at tillering and maturity stages showed lower acceptability score for the Digalu variety followed by the Danda'a variety, indicating higher preferences for the varieties owing to their attribute of resistance to yellow rust, higher vigour, and superior grain yields.

Table 1. Matrix ranking of farmers' preferences for bread wheat varieties at tillering and maturity stages in Gurawa woreda (district) in the highlands of eastern Ethiopia.

Criteria	Tillering stage			
	Madda Walabu	Digalu	Danda'a	Local
Stand uniformity	3	1	2	4
Tillering capacity	3	1	2	4
Stand vigour	2	1	3	4
Leaf greenness	3	1	2	4
Resistance to disease (yellow rust)	3	1	2	4
<i>Acceptability score*</i>	<i>14</i>	<i>5</i>	<i>11</i>	<i>20</i>
	Maturity stage			
	Madda Walabu	Digalu	Danda'a	Local
Resistance to disease (stem rust)	3	1	2	4
Tolerance to sprouting	2	1	3	4
Number of seeds per spike	3	1	2	4
Straw yield	2	1	3	4
Grain yield	3	1	2	4
Seed plumpness	3	1	2	4
Seed weight	3	1	2	4
<i>Acceptability score*</i>	<i>19</i>	<i>7</i>	<i>16</i>	<i>28</i>

\*The lower the acceptability score value in the matrix ranking, the higher farmers' preferences for the variety with respect to the parameter evaluated.

### 3.2. Quantitative Evaluation of Growth and Yield

Analysis of the data indicated that the varieties differed significantly in the number of days required to reach physiological maturity and in the resistance to yellow rust (*Puccinia striiformis* f.sp. *tritici*), which is rampant in the area. Digalu matured latest whilst the local variety matured earliest. The other two varieties exhibited the trait of a medium time of maturity. Thus, in terms of the length of time required in days to reach maturity, the varieties were ranked as Digalu > Danda'a > Madda Walabu > Local (Table 2). The yellow rust severity score revealed that the local variety was very susceptible while Digalu was very resistant to the disease. Madda Walabu and Danda'a were slightly infected by the yellow rust disease. In south-eastern Ethiopia, it was also reported that these two varieties had a slow type of resistance, technically known as Adult Plant Resistance (APR), and not totally immune to the rusts (Solomon and Firdissa, 2012). This means

that the infection is below the threshold level, which would not cause economical yield losses.

The results of the study also indicated that plant height, number of spikes per m<sup>2</sup>, and number of tillers per plant of all the three improved varieties (Madda Walabu, Digalu, and Danda'a) were in statistical parity. However, the local variety had significantly lower values for these growth parameters, indicating its poor performance compared to the improved ones. In line with this result, other studies conducted in the region showed that the local wheat variety required a smaller number of days to reach maturity. In addition, the local variety was reported to have lower number of tillers per plant, with shorter plants than those of the improved varieties (FARC, 2013).

Analysis of the yield-related traits revealed that Madda Walabu produced significantly lower numbers of seed per spikelet and spike than Digalu and Danda'a (Table 3). On the other hand, Digalu and Danda'a produced higher numbers of seeds per spikelet and

spike. However, the numbers of seeds per spikelet and spike produced by Digalu and Danda'a were in statistical parity with those of the local variety. All of the varieties showed non-significant differences in spike length and number of spikelet produced per spike. On the other hand, significantly higher numbers of kernels per spike were produced by Digalu and Danda'a. In south-eastern and northern Ethiopia, higher numbers of seeds per spike were recorded for Digalu and Danda'a compared to the other two varieties (EAAP, 2012).

Variety Digalu produced also a significantly higher biological yield than the other varieties, indicating that production of this variety has an ecological appeal in terms of carbon sequestration and nutrient cycling upon return of the residue into the soil. The result also indicates that the variety has a higher use value the straw as livestock feed compared to the other varieties. On the other hand, Madda Walabu, Danda'a, and the local variety produced biomass yields that were in statistical parity. Other studies also showed that Digalu produced a higher biomass yield than other wheat varieties (EAAPP, 2012).

The improved varieties produced comparable grain yields with each other. However, they produced significantly higher yields than the yield produced by the local variety. Thus, Digalu, Danda'a, and Madda Walabu produced about 54, 49, and 34% higher grain yields than the local variety, in the order mentioned here. On the other hand, Danda'a and Madda Walabu had higher harvest indices, indicating higher dry matter partitioning to the grains. Consistent with this result, other studies conducted in eastern and south-eastern Ethiopia showed that the grain yields obtained from Digalu and Danda'a markedly exceeded the grain yields obtained from other improved and local wheat varieties (FARC, 2013; EAAPP, 2012). Concordant with the results obtained in this study, FARC (2013) reported that, in the East Hararghe Zone, Madda Walabu variety produced a grain yield that was comparable with the grain yields of Digalu and Danda'a although it showed only moderate resistance to rust diseases. Generally, according to production statistics, Digalu and Danda'a produced grain yields that were four-fold higher than the average grain yield commonly obtained by farmers in the region (CSA, 2013).

Table 2. Growth performances and resistance to yellow rust disease of bread wheat (*Triticum aestivum* L.) varieties grown on farmers' fields in Gurawa woreda (district) in the eastern highlands of Ethiopia.

Variety	Yellow rust severity (score: 0-5)	Days to maturity	Plant height (cm)	No. of spikes m <sup>-2</sup>	Spike length (cm)	No. of fertile tillers plant <sup>-1</sup>
Madda Walabu	1.00 <sup>b</sup>	134.50 <sup>c</sup>	102.00 <sup>a</sup>	501.25 <sup>a</sup>	9.38	2.15 <sup>ab</sup>
Digalu	0.00 <sup>d</sup>	141.75 <sup>a</sup>	103.50 <sup>a</sup>	476.25 <sup>a</sup>	7.85	2.60 <sup>a</sup>
Danda'a	0.65 <sup>c</sup>	137.75 <sup>b</sup>	104.70 <sup>a</sup>	477.50 <sup>a</sup>	8.48	2.35 <sup>a</sup>
Local	3.50 <sup>a</sup>	131.25 <sup>d</sup>	86.55 <sup>b</sup>	361.25 <sup>b</sup>	8.13	1.70 <sup>b</sup>
SE ±	0.13	0.64	2.91	14.05	0.45	0.14
LSD	0.34	2.03	9.31	44.93	Ns	0.45
CV (%)	1.46	0.93	5.87	6.19	10.72	12.86

Means followed by the same letters are not significantly different at 5% level of significance according to the Least Significant Difference (LSD) test. \*log transformed

Table 3. Yield and yield-related traits of bread wheat (*Triticum aestivum* L.) varieties grown on farmers' fields at Gurawa woreda (district) in the highlands of eastern Ethiopia

Variety	No. of spikelet spike <sup>-1</sup>	No. of seeds spikelet <sup>-1</sup>	No. of seeds spike <sup>-1</sup>	Biological yield (kg ha <sup>-1</sup> )	Grain yield (kg ha <sup>-1</sup> )	Harvest index
Madda Walabu	18.30	2.96 <sup>b</sup>	54.40 <sup>b</sup>	17125 <sup>b</sup>	6502.50 <sup>a</sup>	0.38 <sup>ab</sup>
Digalu	19.60	3.50 <sup>a</sup>	68.26 <sup>a</sup>	21250 <sup>a</sup>	7432.50 <sup>a</sup>	0.36 <sup>b</sup>
Danda'a	18.65	3.47 <sup>a</sup>	64.55 <sup>a</sup>	16875 <sup>b</sup>	7193.75 <sup>a</sup>	0.43 <sup>a</sup>
Local	18.64	3.25 <sup>ab</sup>	60.40 <sup>ab</sup>	14125 <sup>b</sup>	4835.0 <sup>b</sup>	0.34 <sup>b</sup>
SE ±	0.56	0.12	2.55	1015.72	439.00	0.02
LSD	Ns	0.38	8.17	3249.50	1404.40	0.06
CV (%)	5.96	7.15	8.25	11.71	13.53	10.38

Means followed by the same letters are not significantly different at 5% level of significance according to Least Significant Difference tests.

### 3.3. Pre-Scaling up of Selected Varieties

Based on the results of the participatory agronomic evaluation, the selected varieties (Digalu and Danda'a) along with their improved management practices were

recommended for pre-scaling. The pre-scaling up activities were conducted in 2013 on wider areas by involving Farmers' Research Extension Groups (FREGs), which were upgraded from FRGs. Women

were included (16%) in the process to enhance adoption of the varieties. As part of the pre-scaling up activities, on-spot training was given to experts of offices of agriculture, Development agents (DAs) and Farmers' Research and Extension Groups (FREGs) on wheat production technologies. In the process, more emphasis was given to how technological and technical outputs could be shared among the farmers and with other stakeholders in order to assist further scaling-up. A series of hands-on training were given to the FREGs on improved agronomic practices and seed maintenance techniques. CASCAPE innovators provided technical backstopping to DAs and farmers at the critical stages of activity implementation.

The pre-scaling up activities were conducted in two ways: through direct supply of improved seeds, other inputs and technical supports by the CASCAPE project, and through supply of seeds by FRG members (trial farmers of the previous year) and technical support by CASCAPE. Accordingly, varieties Digalu and Danda'a along with their improved agronomic practices were produced by 115 farmers on the total land area of 14.4 hectares. Feedback collected during the field days showed that farmers were interested in CASCAPE's participatory and step-wise approaches, which addressed their uncertainty about the merit of improved varieties, the critical shortages of seeds of the crop, and enhanced their know-how on improved agronomic practices.

It is believed that the critical shortage of seeds of improved wheat varieties in Ethiopia could be solved through farmer-to-farmer dissemination of seeds since the seed enterprises are not interested in multiplying seeds of new crop varieties that are important for household and national food security (self-pollinated, open pollinated, and vegetatively propagated crops) compared to the profit-oriented hybrid varieties (Dawit, 2010).

The results of this trials ascertained that there is a need for involving the users of agricultural technologies (the farmers) to clear their uncertainties about new technologies. This suggestion is consistent with the finding of Thompson and Scoones (2009) that lack of adoption of improved varieties has been attributed to the linear character of agricultural knowledge and information systems, which do not involve users of the varieties (farmers). In response to this problem, Bishaw and Turner (2008) proposed participatory research and development systems, which put farmers at the centre of the innovation process. The participatory approach places a high value on local knowledge and seeks cooperation between farmers and researchers when designing new technologies or adapting existing ones to local circumstances (Ceccarelli and Grando, 2007; Sperling *et al.*, 2001).

#### 4. Conclusion

The results of the trials have demonstrated that, out of the three improved wheat varieties, two varieties,

namely, Digalu (HAR-3116) and Danda'a were preferred by farmers most on the merits of higher grain yields, resistance to disease, and overall adaptability to the area. The preference for only the two varieties signifies that necessarily not all improved varieties released through the research system could be eagerly adopted by farmers. This exemplifies that appraising existing improved crop varieties through farmers' participatory evaluation processes in each agro-ecology is a prerequisite for adoption and scaling up of production of the crop. In conclusion, the results of this study have revealed that farmer participatory evaluation of new or already existing improved crop varieties is a vital prerequisite for adoption and scaling up of production for enhanced food security and improved farmers' livelihoods in the region.

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## Registration of “*Abdissa and Moti*” Triticale (*X-Triticosecale wittmack*) Varieties

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**Abstract:** Triticale varieties that have pedigree names TCL-76 and TCL-61 and common names “Abdissa and Moti”, respectively, were collected from Kulumsa Agricultural Research Center and evaluated under a regional variety trial for two years and released by Bako Agricultural Research Center. These varieties were evaluated for two consecutive years at Shambo, Gedo and Arjo districts and for one year at Diga district of western Oromia. The varieties performed well and were found to be tolerant to the major diseases (Septoria, leaf, and stem rusts) in the area. Multi environmental trial (MET) analysis indicated that these varieties have better yield and agronomic performances than the standard check (Dilfekar) and are most stable across the years and locations.

**Keywords:** Triticosecale wittmack; Pedigree

### 1. Introduction

Triticale (*X-Triticosecale Wittmack*) is a man-made crop developed by crossing wheat (*Triticum turgidum* or *triticum aestivum*) with rye (*Secale cereale*). It is an amphidiploid crop with  $2n = 56$  chromosomes (42 from wheat and 14 from rye) (Allard, 1960). It can adapt to a wide range of soil conditions ranging from sandy to clay soil type and also exhibited better performance under acidic and degraded soils compared to many other cereals (MARD, 2006)

### 2. Agronomic and Morphological Characteristics

Abdissa and Moti are characterized by their awn and awnless characteristics respectively. Easy threshing ability is one of the best characteristics of the Moti variety. When compared to other triticale varieties, the seed of Moti is larger than that of Abdissa. Moti has also bigger spike than Abdissa. The seed color of both varieties is creamy white (Table 4) Both varieties have an erect growth habit and are resistant to lodging.

### 3. Yield Performance

The two varieties were evaluated against thirteen genotypes and one standard check variety, Dilfekar, at Shambo, Gedo, Arjo and Diga districts of western Oromia for two consecutive years (2011 and 2012) for their adaptability and yield performance. Among the tested genotypes Abdissa and Moti performed better with grain yields of 5.6 and 4.7 t ha<sup>-1</sup>, respectively, than the standard check (4.6 t ha<sup>-1</sup>) (Table 2). Moreover, these varieties were evaluated by farmers following a participatory approach and TCL-61 (Moti) variety was selected due to its easy threshing ability and bigger spike length since spike length is mainly associated with the number of seed per spike and the size of the seed.

### 4. Stability Performance / Adaptability

Yield stability for fifteen *Triticale* genotypes and the standard check was studied for stability across environments. According to Ebrehart and Russell (1996), the genotype with higher mean grain yields, unity regression coefficient ( $b_i$ ) and the value for squared deviation from regression ( $s^2_{di}$ ) approaching zero are stable and widely adaptable. Accordingly, genotype TCL-76 (Abdissa) had the highest grain yield and the regression coefficient ( $b_i$ ) was significantly higher than unity. This implied that this genotype is highly responsive to the change in environment and could be recommended for western Oromia and similar agro-ecologies of the country with appropriate agronomic practices. However TCL-61 (Moti) had comparable grain yield with the standard check and the regression coefficient ( $b_i$ ) was slightly lower than unity (0.976).

### 5. Disease Reaction

Abdissa and Moti are resistant to the predominant diseases; Septoria (*Septoria tritici*), leaf (*Puccinia recondata*) and stem rust (*Puccinia graminis*) of the area.

### 6. Conclusion

Abdissa (TCL-76) produced the highest mean grain yield but the regression coefficient ( $b_i$ ) was significantly higher than unity. This indicates that this variety is highly responsive to the change in environment and recommended for specific locations of western parts of the country with appropriate agronomic practices. Even though the regression coefficient ( $b_i$ ) of Moti (TCL-61) was slightly lower than unity. It is an awnless triticale variety which has long spikes, and better disease reactions across the tested environments. Though this variety has comparable yield with the standard check, it is preferred by farmers due to its easy threshing, and is recommended for Shambo, Arjo,

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Gedo and areas with similar agro-ecological conditions in the country.

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Table 1. Phenological, growth, and agronomic performances of the varieties in 2010 and 2011 G.C at Shambo, Gedo and Arjo sub-sites.

Acc	Days to heading							Days to maturity						
	2010			2011			Mean	2010			2011			Mean
	Gedo	Shambo	Arjo	Gedo	Shambo	Arjo		Gedo	Shambo	Gedo	Shambo	Arjo		
TCL-76	66.3	63.3	73.0	64.5	64.5	72.0	67.3	135	127.0	154.8	133.3	141.8	138.4	
TCL-61	66.5	63.3	75.3	62.3	66.3	74.2	68.0	134	125.3	154.0	131.5	143.5	137.7	
Dilfikir	60.8	59.8	79.0	61.0	60.2	79.0	66.6	136	124.0	154.0	132.8	144.0	138.2	
Mean	64.5	62.1	75.8	62.6	63.7	75.1		135.0	125.4	154.3	132.5	143.1		
CV	2.71	2.1	3.6	2.80	2.4	2.8		1.67	0.86	0.64	1.0	1.6		
LSD (5%)	2.0	1.5	3.2	2.2	1.8	3.3		3.20	1.55	1.2	1.5	2.8		
F-value	**	**	NS	**	**	**		*	**	NS	**	**		

Where, CV = Coefficient of variation; \* = Significant at 5% level of significance; \*\* = Significant at 1% level of significance; LSD = Least Significant Test at 5% level of significance; TKW = Thousand kernel weight; ACC= Accession, 1000KW= Thousand kernel weight

Table 2. Phenological, growth, and agronomic performances of the varieties in 2011 G.C at Shambo, Gedo and Arjo sub sites.

Acc	Plant height (cm)							Grain yield (t ha <sup>-1</sup> )							Mean
	2010			2011			Mean	2010			2011				
	GD	SH	AR	GD	SH	AR		GD	SH	AR	GD	SH	AR	DG	
TCL-76	101	109.8	96.8	114.0	111.8	101.3	106	2.365	6.580	6.288	7.62	7.18	5.68	3.99	5.67
TCL-61	110	112.3	90.5	110.0	107.0	101.0	105	2.775	5.050	4.524	6.48	6.03	5.61	2.86	4.76
Dilfikir	110	114.0	94.5	112.0	109.0	97.8	106	2.664	5.240	5.59	5.82	6.74	5.14	1.50	4.66
Mean	107.	112.0	93.9	112.0	109.3	100.0		2.60	5.62	5.45	6.64	6.65	5.48	2.78	
CV	4.39	3.68	6.08	3.45	5.3	7.23		NS	*	*	17.2	16.4	18.6		
LSD (5%)	7.13	6.42	8.17	4.9	7.1	9.2		NS	*	*	13.54	11.58	15.66		
F-value	**	**	*	**	**	**		NS	*	*	NS	*		NS	

Where, GD = Gedo; SH= Shambo; AR=Arjo; DG=Diga

Table 3. Analysis of variance for Additive Mean Effect and Multiple Interactions (AMMI).

Source	Df	MS	%G x E interaction explained
Environments	6	105.87**	
Reps within Env.	21	4.29	
Genotype	15	2.30*	
Genotype x Env.	90	1.67**	
IPCA 1	20	3.30**	43.86
Total	447		
Residual	315	1.09	

Table 4. General characteristics of the varieties.

Characteristics	Varieties	
	Abdissa (TCL-76)	Moti (TCL-61)
<b>Pedigree</b>	ARDI/GNU//2*FAHAD_1/4/ERIZO_6/NIMIR	BULL_10/MANATI_1//FARAS/CMH84.414
Adaptation area:		
Altitude requirement (masl)	1800-2700	1800-2700
Rainfall requirement (mm)	>600mm	>600mm
Fertilizer rate		
DAP (kg ha <sup>-1</sup> )	100	100
Urea (kg ha <sup>-1</sup> )	50	50
Fertilizer application method and time		
DAP	Row application at planting	Row application at planting
Urea	Row application at 3-4 leaf stage	Row application at 3-4 leaf stage
Seeding rate (kg ha <sup>-1</sup> )	125-130	125-130
Spacing (cm)		
Between rows	20 cm	20 cm
Planting date	Late June – Early July	Late June – Early July
Days to heading	63-73	63-75
Days to maturity	127-154	127-154
1000 seed weight (g)	44.2	40.4
Plant height (cm)	97-114	125-154
Awn presence	Present	Awn less
Seed color	White Cream	White Cream
Growth habit	Erect	Erect
Grain yield (q/ha)		
On farmers field	40-50	38-45
On-station	44-62	40-53



## Registration of a Faba Bean variety named *Gora*

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**Abstract:** A faba bean (*Vicia faba* L.) variety named *Gora* with the pedigree designation of EK01024-1-2 has been released by Kulumsa agricultural research center in Ethiopia. The variety is best adapted to altitudes ranging between 1900-to-2800 meters above sea level in the country. The variety was developed through hybridization between an adapted genotype 'EH91026-8-2' with bean pure line 'BPL44-1'. It has been tested at Kulumsa, Asassa, Bokoji, Koffale, Holetta, Adadi, Jeldu, Haramaya, Adet, Sinana and Shambu from 2009 to 2011 main cropping seasons. The variety is mainly characterized by a heavier seed (938 g/1000 seeds) than seeds any other faba bean varieties released to date in the country. The seed weight of this variety is 17% heavier than the seed weight of the variety used as the standard check. Based on most stability parameters, *Gora* showed relatively better grain yield performance and stability across a range of environments and years than the standard checks *Moti* and *Gebelcho*. This variety is moderately resistant to the major faba diseases such as chocolate spot and rust, and could be cultivated across a number of locations in the mid and high altitude areas of Ethiopia for increasing productivity of the crop.

**Keywords:** Disease resistance; Grain yield; National yield trail; Preliminary variety trial, Seed size; *Vicia faba* L.,

### 1. Introduction

Faba bean (*Vicia faba* L.) is the most important pulse crop in terms of both area coverage and volume of annual production in Ethiopia. Currently, it occupies about 574,061 hectares of land with an annual national production of 943,964.2 tons, with a productivity of 1.64 tons ha<sup>-1</sup> (CSA, 2013). Ethiopia is the first producer of faba bean in Africa and the second in the world next to the Peoples Republic of China (Mussa and Gemechu, 2006). The crop is mainly cultivated in mid and high altitude areas, with an elevation ranging from 1800-3000 meters above sea level (Mussa and Gemechu, 2006). The inception of faba bean breeding in Ethiopia was in the 1950's with the establishment of Arsi Rural Development Unit (ARDU) followed by Alemaya (now Haramaya) College of Agriculture. The main objectives of faba bean breeding in Ethiopia are to improve its productivity through developing and promoting improved cultivars with high and stable yield, and resistant/tolerant to major biotic and abiotic stresses (Gemechu *et al.*, 2006). A special focus has been given to improve grain yield, and diseases and water-logging resistance or tolerance. Very recently, considerable attention has been paid to develop large-seeded faba bean varieties to meet the demand of the export-market for seed quality since large-sized seeds are preferred by consumers in the local market and fetch premium prices in the international market.

### 2. Origin and Varietal Evaluation

The earlier adapted faba bean variety 'EH91026-8-2', which was selected from the last stage of variety trial, was crossed with bean pure line (BPL44-1) introduced from ICARDA. The crossing was done at Kulumsa Agricultural Research Center during 2001 cropping season. As faba bean is partially allogamous, screen houses were routinely used in the early generations, i.e., F<sub>1</sub>, F<sub>2</sub>, F<sub>3</sub> and F<sub>4</sub>, of a breeding cycle to prevent bees from causing cross-pollination. During these phases, selection for traits with high heritability such as seed size, grain yielding ability, plant habit, time of flowering and resistance to major diseases such as chocolate spot and rust were undertaken. Thirty-six elite individual lines selected from the F<sub>5</sub> generation were promoted and evaluated for yielding ability, large seed size, disease reaction and stability at in a preliminary variety trial (PVT) conducted during the 2008 cropping season at multi-locations. From this trial, 14 promising genotypes were promoted and evaluated in a national yield trial (NVT) along with two recently released standard checks '*Moti* and *Gebelcho*' at multi-locations. The locations where the trials were conducted included Kulumsa, Asassa, Bokoji, Koffale, Holetta, Adadi, Jeldu, Haramaya, Adet, Sinana and Shambu from 2009 to 2011 main cropping seasons. The trials were replicated four times per location. Finally, EK01024-1-2 and EK01001-5-1 were selected as the most promising candidate varieties and evaluated along with two best standard checks on 10 m x 10 m plots by the national variety release technical committee at 7

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locations, each one on-station and two on-farm fields during the 2012 cropping season. Eventually, EK01024-1-2 was recommended for commercial production and named *Gora*.

### 3. Varietal Characteristics

The newly released faba bean variety '*Gora*' is characterized by an indeterminate growth habit. Its flower color is white with black spots. The seed coat and cotyledon colors are pale green and ceramic, respectively. The average number of days required by the variety to reach its 50% flowering and 95% physiological maturity were 59 and 139, respectively, with the average plant height being 131 cm (Table 1). The average number of pods per plant is 10.3 (Table 1). The appropriate planting date for this variety would range from early June to early July. For a better harvest,

the variety must receive 46 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and 18 kg ha<sup>-1</sup> N at sowing.

### 4. Yield and Quality Performance

The released variety '*Gora*' is mainly characterized by a heavier seed than the seeds of other hitherto released faba bean varieties in the country, which averages 938 g per 1000 seeds. The seed of this newly released variety has weight advantages of 17% and 15% over the standard checks *Moti* and *Gebelcho*, respectively. In addition to its seed size advantage, the average grain yield of the newly released variety, combined over locations and years, exceeded the average yield of *Moti* by 4.3% and that of *Gebelcho* by 10.7% (Table 1). The data on quality traits indicated in Table 1 show that the released variety '*Gora*' has a comparable quality with those of the standard checks.

Table 1. Mean grain yield, agronomic traits, quality parameters and disease reaction of '*Gora*' among two standard checks tested in 16 environments during 2009/2010-2011/2012 cropping seasons.

ENTRY	Agronomic traits				TSW (g)	Grain yield (kg/ha)	Disease reaction		Quality parameters	
	DTF	DTM	PHT (cm)	NPPP			ChS	Rust	ACP (%)	Soak-ability (%)
Moti (Check-1)	58	137	132	12.16	801	3657	30.0	27.7	22	90.2
Gebelcho (Check-2)	61	140	130	11.15	814	3447	28.5	25.8	23	83.0
Gora (EK01024-1-2)	59	139	131	10.28	938	3815	28.6	26.7	24	92.0

DTF = Days to 50% flowering; DTM = Days to 95% physiological maturity; PHT = Plant height; NPPP = Number of pods per plant; TSW = 1000 seed weight; ChS = Chocolate spot; ACP = Average crude protein.

### 5. Performance Stability and Adaptation Domain

The variety '*Gora*' was released for the mid-to-high altitude agro-ecologies of the country receiving 700-to-1100 mm average annual rainfall. It is well adapted to an altitude range of 1900 to 2800 meters above sea level such as Kulumsa, Holleta, Bokeji, Asassa, Jeldu, Kofele, Sinana, Adet, Haramaya and similar agro-ecologies. Based on most stability parameters, '*Gora*' showed relatively better performance stability across a range of environments. Most of the univariate parametric methods such as  $bi$ ,  $S^2di$ ,  $a$ ,  $\lambda$ ,  $Pi$ ,  $ASV$ , and the non-parametric method  $RS$  were identified '*Gora*' as the most desirable genotype (Table 2). According to Lin *et al.*, (1986), both Francis and Kannenberg's (1978) coefficient of variability ( $CV$ ) and environmental variance ( $EV$ ), and those methods which measure

phenotypic stability based on the amount of sum of squares contributed by each genotype into the interaction effect, for example,  $P^{59}$ ,  $\sigma_i^2$  and  $W_i$  are classified into Type I stability concepts. Genotypic stability according to these parameters is heritable and its genetic mode is additive and consistent. Though having both high yield and Type I stability concept occurs rarely in multi-location trials (Karimzadeh *et al.*, 2012), it was evident that this faba bean variety demonstrated smaller values for these stability methods and was identified as the most stable genotype possessing Type I stability concept (Table 2). Furthermore, based on the stratified ranking technique of Fox *et al.* (1990) of the top three ( $FT3$ ) parameters, *Gora* is a top yielding genotype which was ranked in the top third of the entries in 61.5% of the test environments (Table 2).

Table 2. Parametric and non-parametric stability statistics of ‘Gora’ variety among two standard checks tested across 16 environments during 2009/2010 – 2011/2012 cropping seasons.

Varieties	Parametric and non-parametric stability methods												
	Bi	S <sup>2</sup> di	Wi	CVi	EV	$\sigma^2$	$\alpha$	$\lambda$	Pi	ASV	P <sup>59</sup>	FT3	RS
Moti (Check-1)	1.12	0.12	2.41	29.54	1.25	0.22	0.13	2.25	0.19	0.99	0.17	61.54	18
Gebelcho (Check-2)	1.13	0.06	1.84	29.9	1.21	0.17	0.13	1.68	0.17	<u>0.47</u>	0.14	46.15	19
Gora (EK01024-1-2)	<u>1.03</u>	<u>0.01</u>	<u>1.09</u>	<u>25.49</u>	<u>0.98</u>	<u>0.10</u>	<u>0.03</u>	<u>1.09</u>	<u>0.11</u>	0.56	<u>0.11</u>	<u>61.54</u>	<u>8</u>

Bi = Regression coefficient; S<sup>2</sup>di = deviation from regression; Wi = Wricks's ecoralance;  $\sigma^2$  = Shukla's stability variance; CVi = Coefficient of variation; EV = Environmental variance;  $\alpha$  and  $\lambda$  = Tai's alpha and lambda; P<sup>59</sup> = Plaisted and Peterson's stability parameter; Pi = Lin and Binn's superiority index; ASV = AMMI Stability Value; RS = Kang's rank sum; FT3 = Number of sites at which the genotype occurred in the top third of the ranks. Underlined are the most stable..

## 6. Reaction to Major Diseases

Developing resistant or tolerant varieties to major diseases such as chocolate spot (*Botrytis fabae*) and rust (*Uromyces viciae-fabae*) is among the major objectives of the national faba bean breeding program. Chocolate spot and rust scores based on (1-9) scale were converted to pre-transformed percentage values, which were then used to determine the reaction of the released variety ‘Gora’ to major diseases (Little and Hills, 1978). Accordingly, the released variety ‘Gora’ showed an average reaction of 28.6 and 26.7% for chocolate spot and rust, respectively (Table 1), and is characterized as moderately resistant to these major diseases.

## 7. Variety Maintenance

The breeder and foundation seed will be maintained by Kulumsa Agricultural Research Center.

## 8. Conclusion

Grain yield is the primary trait of interest and a prime objective in faba bean breeding programs for many decades. However, also seed size has received a special attention recently. This is also what is happening at international and national levels in response to the current move to meet the export-market demand for seed quality particularly for the development of large-sized seeds that fetch high prices in the world market. Regardless of this, only few varieties that combine both high yield with large seed sizes have been released since the inception of faba bean breeding program in the country. The current variety, *Gora*, has almost 80% and 15% seed size advantages over the widely cultivated small seeded faba bean varieties, *Degaga* and *CS20DK*, and the large-seeded variety *Gebelcho* with comparable seed yield productivity, respectively. Therefore, wide cultivation of *Gora* variety will boost productivity and marketability of the crop and improve farmers' income.

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